

***Fraxinus angustifolia* Vahl as a valuable species in  
riparian rehabilitation projects**

**From annual growth to habitat preference of narrow-leaved ash  
in Southern Portugal**

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## ABSTRACT

Mediterranean riverine systems show distinct features due to their marked seasonal pattern. Species present in these environments developed specific functional adaptations to cope with this variability linked to events like floods, droughts and geomorphic natural disturbances. Widespread river and floodplain alteration across Mediterranean regions, turned restoration and rehabilitation of riparian corridors a subject of increasing interest during the last decade. Yet, the complex dynamics of riverine Mediterranean systems and their vegetation communities' interaction is still under study. Improving our understanding on species functional responses to environmental variability will contribute to successful restoration/rehabilitation through more ecologically and economically efficient projects.

It was hypothesized that tree growth is affected by water availability and that trees have higher growth near the water channel and downstream from the river source.

In this dissertation, dendrochronology was used to study past tree growth in *Fraxinus angustifolia* Vahl trees from Odelouca river, in Algarve. Annual growth was analyzed in groups differing in their relative position to the active river channel and distance to catchment source, and analyzed with hydrological, climatic and edaphic factors.

Tree position relative to active river channel showed to have the highest impact on mean raw tree growth during the last four decades, followed by soil C/N ratio. High minimum temperature during autumn extended the growing season, a trend associated with climate change and temperature rise in the last three decades in the Mediterranean. Additionally, discharges during spring created favorable conditions when tree growth was more intense.

Establishing optimal growth position and ecological conditions for riverine species is important to define principles and guidelines for restoration and rehabilitation projects. Also, global change plays an important role in modulating future responses of vegetation under a changing climate, so understanding past responses can provide important clues for adaptive management.

Key Words: Dendrochronology, *Fraxinus angustifolia*, Mediterranean ecosystems, riparian restoration



## RESUMO

Os sistemas ripários Mediterrâneos são muito particulares devido ao seu padrão sazonal de seca e cheia. As espécies presentes nestes ambientes desenvolveram adaptações funcionais específicas de forma a resistir à variabilidade de condições, como cheias, secas e alterações geomórficas naturais. As alterações nos rios e vales de cheia ao longo das regiões mediterrâneas tornou o restauro e a requalificação de galerias ripícolas um tema cada vez mais abordado, no entanto devido à complexidade destes sistemas este é um tema ainda em estudo. É por isso importante aumentar o conhecimento sobre as respostas funcionais das árvores à variabilidade ambiental, para apresentar projectos de restauro e requalificação mais eficientes a nível ecológico e económico.

As hipóteses propostas são: o crescimento anual da árvore é afectado pela disponibilidade hídrica e as árvores com posições mais perto do canal do rio e mais a jusante têm crescimentos maiores. Nesta dissertação foram usados métodos de dendrocronologia para estudar o crescimento passado de exemplares de *Fraxinus angustifolia* Vahl da Ribeira de Odelouca, no Algarve.

A posição da árvore relativamente ao canal activo mostrou ser o factor com maior impacto no crescimento das últimas quatro décadas, seguido do rácio Carbono/Azoto no solo. O aumento da temperatura mínima no Outono prolongou a estação de crescimento, um efeito associado às alterações climáticas e aumento da temperatura na região Mediterrânea nas últimas décadas. Adicionalmente, os valores de caudal de Primavera mostraram criar condições favoráveis ao crescimento, na época em que este é mais intenso.

De forma a definir princípios e linhas de orientação para projectos de restauro e requalificação é importante estabelecer condições ecológicas óptimas de crescimento. As alterações globais têm mudado as respostas da vegetação ao longo do tempo e por isso é importante perceber quais as respostas passadas para ser possível criar melhores formas de gestão futuras.

Palavras-chave: Dendrocronologia, *Fraxinus angustifolia*, ecossistemas mediterrâneos, restauro de galeria ripária



## RESUMO ALARGADO

As árvores de galerias ripícolas de sistemas mediterrâneos têm de se adaptar a situações específicas deste habitat e clima e embora tenham acesso privilegiado a uma fonte de água, as estações seca e quente (Verão) e fria e húmida (Inverno) afetam o seu crescimento anual. As suas estratégias de reprodução e sobrevivência precisam de estar em sintonia com as características de cada rio. Concretamente, cada sistema lótico de água doce tem as suas características específicas de regime de caudal, leito de cheia, período de retorno, entre outros. Estas características tornam necessário o conhecimento da ecologia das espécies presentes e das suas respostas funcionais, tais como o crescimento, com vista ao seu restauro.

Cada caso de restauro ou requalificação deverá ter em atenção a situação particular de cada sistema dentro da bacia hidrográfica bem como as condições existentes antes da perturbação para que estas sejam repostas após a intervenção humana. Para as operações de restauro ou requalificação são usadas espécies que melhor se poderão adaptar ao local perturbado mas que também correspondam o máximo possível ao ambiente natural. É por isso importante entender o comportamento das espécies vegetais constituintes da galeria ripícola de forma a conhecer como estas espécies poderão ser usadas para obter o máximo sucesso possível no seu crescimento e do corredor ripário nos anos seguintes à operação.

As respostas da vegetação às condições ambientais têm-se alterado ao longo das últimas décadas devido ao aquecimento global, tendo vindo a registar-se alterações na fenologia das espécies e nas suas condições de crescimento. Tal acrescenta uma nova dinâmica ao restauro, pois deverá ter em conta o estado natural, mas as espécies “engenheiras” deverão ao mesmo tempo ser resistentes às alterações de que atualmente os sistemas estão a ser sujeitos.

O estudo do crescimento histórico anual de uma árvore pode dar indicações importantes acerca do tipo de resposta de crescimento a fatores de stress presentes no ambiente, permitindo transpor essa informação para o uso presente das espécies em ações de restauro ou requalificação.

O objetivo deste trabalho passa por perceber quais os principais fatores limitantes e favoráveis do crescimento de *Fraxinus angustifolia* Vahl, uma espécie arbórea ripícola de clima mediterrâneo na ribeira de Odelouca, no Algarve. Devido aos resultados de estudos anteriores em espécies com habitats semelhantes espera-se que o crescimento seja negativamente afetado pela diminuição de caudal nos meses de Verão e aumento do mesmo nos de Inverno. Espera-se também que as árvores situadas mais longe do canal ativo e mais perto da nascente apresentem um menor crescimento radial.

Neste estudo foram usadas técnicas de dendrocronologia para datar os anéis de crescimento anual de cada indivíduo, depois, recorrendo a ferramentas estatísticas várias variáveis

independentes foram testadas como fatores significantes nos modelos de crescimento dos indivíduos.

Esta dissertação segue o formato de um trabalho científico. É iniciada com uma introdução onde são explicados os objetivos do trabalho e a sua estrutura geral, seguido de uma revisão bibliográfica com os conceitos mais importantes e principais informações já disponíveis sobre o tema. A revisão bibliográfica encontra-se dividida em quatro partes: os sistemas ripícolas mediterrâneos e restauro do corredor ripícola; dendrocronologia; efeitos de fatores ambientais no crescimento de árvores e a espécie em questão. Seguido pelo terceiro capítulo onde são apresentados os métodos e materiais usados, o local de estudo assim como a recolha de dados climáticos, hidrológicos, edáficos e de larguras de anel de crescimento. O quarto capítulo apresenta os dados usados nas análises e os resultados de cada análise, que incluem correlações de Pearson, cálculo de cronologias, idade cambial, área de incremento basal e modelos lineares mistos. A discussão dos resultados obtidos é feita no capítulo quinto juntamente com críticas relativamente aos dados usados e as suas limitações. A conclusão, exposta no sexto capítulo, pretende fazer um resumo geral das principais descobertas deste estudo assim como evidenciar futuras questões que necessitam de ser abordadas e representam falhas atuais no conhecimento.

Para efeitos de análise, comparação de dados e interpretação de resultados, as árvores foram agrupadas de acordo com a distância e altura ao canal activo do rio (formando os grupos “perto” e “longe”) e tendo em conta a sua localização ao longo do curso principal e relativamente à ribeira de Benafátima que desagua a meio do troço analisado (formando os grupos “montante” e “jusante”).

Para o estudo dos anéis de crescimento foram usados métodos de dendrocronologia, que permite associar a cada anel um ano específico de crescimento, através de datação cruzada. A análise dos dados em bruto permitiu obter cronologias para o conjunto das amostras recolhidas e para os grupos de amostras formados considerando as características de posição na ribeira (montante e jusante) e distância ao canal ativo da ribeira (perto e longe). Os modelos lineares estudaram a resposta do crescimento para vários períodos de anos, tendo como variáveis explicativas a relação carbono/azoto e fósforo presentes no solo, altura da árvore à água e percentagem da fração de limo e argila no solo.

A cronologia permitiu uma visualização da tendência bruta anual de crescimento para todos os grupos, e uma tendência em que é retirado a influência do crescimento do indivíduo. A comparação entre grupos foi também realizada através da idade cambial, permitindo conhecer as diferenças de crescimento entre grupos nos primeiros anos de crescimento, e de incremento da área basal. O uso de modelos lineares permitiu o uso de variáveis locais a nível da árvore, como os nutrientes do solo, posição da árvore relativamente à distância ao canal ativo da ribeira e granulometria.

Os resultados das análises evidenciam tendências claras no crescimento anual da árvore preferencialmente nos grupos mais longe do canal ativo da ribeira e no grupo a jusante. O crescimento maior no grupo a jusante poderá ser explicado pela maior disponibilidade de água, principalmente devido ao afluente ribeira de Benafátima, que aumenta o caudal a jusante.

Os modelos lineares suportam a evidência anterior ao indicarem uma forte resposta positiva da largura do anel ao aumento da altura da árvore à ribeira durante o período de anos mais longo utilizado para estudo (de 1967 a 2009) e também de períodos de anos mais recentes (de 2005 a 2009 e de 2000 a 2009). Tal como esperado o caudal tem uma correlação positiva significativa com os meses de Verão em que o período de crescimento cambial é mais activo, sendo que estas correlações estão mais presentes em todos os grupos (perto e longe do canal ativo do rio e montante) menos no grupo a jusante. Correlações positivas foram também encontradas para a temperatura mínima, nomeadamente no mês de Setembro, tal indica que o aumento da temperatura mínima potencia o crescimento no mês de Setembro. O aumento da temperatura mínima permite à árvore prolongar o seu período normal de crescimento devido à manutenção das temperaturas e outras condições necessárias para continuar o seu crescimento, nomeadamente o número de horas de sol. A mesma resposta foi obtida para a temperatura média para o mês de Setembro. A precipitação teve resultados positivos importantes nos meses de Janeiro, Fevereiro e Abril, principalmente nos grupos a jusante, precisamente antes do início da estação de crescimento. A temperatura máxima não teve qualquer efeito no crescimento anual das árvores estudadas.

A compreensão de condições favoráveis ao crescimento do freixo em galerias ripícolas permite a aplicação desta espécie em situações de restauro com melhores resultados e mais semelhantes ao habitat natural. As alterações climáticas que continuarão a ocorrer têm alterado o funcionamento dos ecossistemas e a resposta desta espécie elucida qual o seu papel futuro em respostas de crescimento assim como, qual a possível resposta de outras árvores da galeria ripícola com posições e estratégias semelhantes ao freixo.

Este caso de estudo foi realizado na ribeira de Odelouca, uma ribeira torrencial mediterrânea cujo regime hidrológico era considerado como mantido no seu estado natural, no momento de amostragem. Devido à crescente alteração do regime natural dos rios já não é comum encontrar rios ou ribeiras naturais onde seja possível realizar este tipo de estudo, por isso este caso torna-se uma das poucas referências possíveis a nível nacional do tipo de resposta do crescimento desta espécie.

Mais trabalho deverá ser feito no estudo da resposta desta espécie às condicionantes ambientais, nomeadamente em anos específicos de stress ambiental, como cheias e secas. A disponibilidade dos nutrientes no solo revelou-se um importante fator para o aumento do crescimento da árvore e por isso é importante um estudo futuro sobre o efeito de outros nutrientes no crescimento, especialmente em anos de diminuição de disponibilidade de água e portanto a árvore deverá arranjar uma estratégia de crescimento específica.



O delineamento de limites de distância mínimos e máximos em altura á água para a qual existe um crescimento maior ou menor poderá ajudar em situações futuras de tomadas de decisão ao nível de projetos de restauro ou requalificação.

Com este trabalho espera-se ter conseguido fornecer informação nova para melhor compreender o funcionamento desta espécie ripícola mediterrânea e como usá-la em combinação com outras espécies em restauro e requalificação ripária.

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## **LIST OF ABBREVIATIONS**

WFD: Water Framework Directive

BAI: Basal Area Increment

PY: Pointer Years

GLMM: Generalized Linear Mixed Models

EPS: Expressed Population Signal

VIF: Variance Inflation Factor



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## Author's Declaration

A scientific paper is in preparation with relevant results of this study, to be submitted to the journal *Dendrochronologia*:

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# 1. INTRODUCTION

## 1.1 Structure

This dissertation is structured in 6 chapters. The present chapter provides a presentation of the goals and structure, the second presents a bibliographic revision showing state-of-the art across themes important to this study. It includes a description of Mediterranean riverine systems and the concepts involving riparian corridor restoration followed by an explanation of the method of dendrochronology. Then, an overview of the main discoveries by population and work groups and individual tree is given.

The third chapter corresponds to material and methods, introducing the study site, sampling of wood and soil samples and the statistical procedures used for all analysis done on the tree and soil samples.

Results are presented in the fourth chapter, first with a view of the growth curves and responses for all tree groups and after the specific population response to several factors.

Discussion follows in the fifth chapter presenting empirical responses from the results, their connections with riverine tree dynamics and a general trend for tree growth found within the results of the analysis. A review of problems concerning data gathering for this study is also addressed.

Finally, the conclusion shows the objectives achieved by this work and a synthesis of the main findings on the tree growth trends discussed in the previous chapter. Future possible works are presented concerning this species and other riparian species.

## 1.2 Goals

The main goal of this study was to understand whether *Fraxinus angustifolia* Vahl had any growth tendencies related with climatic and/or hydrological data, position to river (height, distance to active channel and distance to source) or soil properties.

The species studied in this work is present in a riverine Mediterranean system and so the main hypothesis proposed is that tree-growth is affected by water availability, mainly in years of drought and in summer, when climatic conditions are severe and water scarcity is frequent. It is also hypothesized that trees have a higher growth near to the water channel and downstream from the river source as drainage area increases. Soil nutrient availability might have an important impact on tree growth, but mainly on years where there is water in abundance, and the limiting factor would be the concentration of nutrients in the soil.

It is intended that the results from this study shed light on the role of this species in riverine restoration or rehabilitation in Mediterranean climate regions, as well as a better understanding of the impacts of climate change on tree growth response. This intends to fill a gap of how climatic, edaphic and hydrological stressors such as resources availability might affect riparian species life history, improving the response predictors of vegetation communities (Stella et al. 2013).

## **2. BIBLIOGRAPHIC REVISION**

### **2.1 Mediterranean Riverine Systems and Riparian Corridor Restoration**

The transition space between river and terrestrial systems is called riparian corridor, in which important processes of sedimentation, periodic flood and erosion take place. The woody formations in riparian corridors have many important functions in the riverine ecosystem, acting as ecological filters improving water quality, contributing to riverbank stabilization, providing nutrients to the surrounding soil and water, functioning as habitat to wildlife and fish, increasing stream shading which regulates the habitats for other organisms such as aquatic plants and improving habitat regional biodiversity (Henry and Amoros 1995).

Mediterranean riverine systems are resource-rich environments occurring in water limited landscapes of four continents, however, occupying a reduced proportion of the total landscape. This makes these systems an interesting study case of particular natural conditions to which several different species from different origins need to adapt. Mediterranean ecosystems have a distinct cool and wet season followed by a warm and dry season therefore are influenced by a sequence of regular and extreme flooding and drying periods (Gasith and Resh 1999).

In Portugal, even in hot and dry summers some riverine areas are the only ecosystems which can maintain for some period humidity conditions, which favor the species in these environments (Fabião and Fabião 2007). In Southern Portugal, river streams in Algarve, like Odelouca, represent a good example of Mediterranean riverine hydrological cycles. In wet season the river flows and floods are common, in dry season the river has a period of 1 to 4 months of water scarcity and in some parts loses river connectivity.

The natural integrity of such an environment depends on several ecological conditions to its best functioning (Poff et al. 1997). The hydrological pattern described above influences development and function of riparian vegetation in combination with many other conditions, like geomorphological dynamics. Yet, rivers natural integrity is currently affected by anthropic activity. The rich floodplain soils transformed for agriculture, the establishment of industrial units close to rivers and river regulation, among many other actions have degraded rivers and the systems connected to them, like riparian corridors.

By the Law nº 19/2014, “As Bases da Política do Ambiente” (AR 2014) which addresses environment in Portugal, every citizen has the right to a humane and ecologically balanced environment as well as the duty to promote the improvement of the quality of life. In order to achieve this goal, there must be prevention against degrading effects and participation of all social groups (national and international). Also, it states that measures should be taken to limit the degradation process and recover areas where such has happened already. In this law inland surface waters are included, as well as animals and plants. Besides protecting ecological processes important for the ecosystems, indigenous fauna and flora is also put under policy protection and recommendations to rational and sustainable water use are made. This

document shows what are the baselines of environmental policy that all should follow, not giving however, specific measures to attain each of the basic principles.

Applying this law to riparian ecosystems, it simply states that any further degradation should be prevented and areas very damaged should be recovered.

The EU Water Framework Directive (WFD), which Portugal is bound to comply, has as one of his purposes to protect inland water masses, so that there will be protection of the water quality (ecologically through biological, physicochemical and hydro morphological elements) and no further deterioration. For that quality to be improved and/or maintained the framework must be applied to water surfaces and the territories contributing to its quality, such as riparian corridors (EU 2000).

The Convention for Biological Diversity established several targets to achieve, like Aichi targets for Restoration (<https://www.cbd.int/sp/targets/>) and EU Biodiversity Strategy 2020 ([http://ec.europa.eu/environment/nature/biodiversity/strategy/index\\_en.htm](http://ec.europa.eu/environment/nature/biodiversity/strategy/index_en.htm)), which would benefit from a deeper knowledge of riparian ecosystems in order to halt the loss of biodiversity and ecosystem services in the EU.

Restoration and rehabilitation are different concepts in ecological systems. Restoration implies the return of the system to its physical, chemical and biological natural parameters before human disturbance (figure 1). However, the lack of references for many river types and the fact that river systems follow complex trajectories frequently making it impossible to return to a previous state (Dufour and Piégay 2009) makes it extremely difficult to achieve this purpose completely (Henry and Amoros 1995). Therefore, most of the times it is used the concept of rehabilitation, that is to retrieve the structure and functions of the river as near as possible to its natural conditions (Melorose et al. 2009).

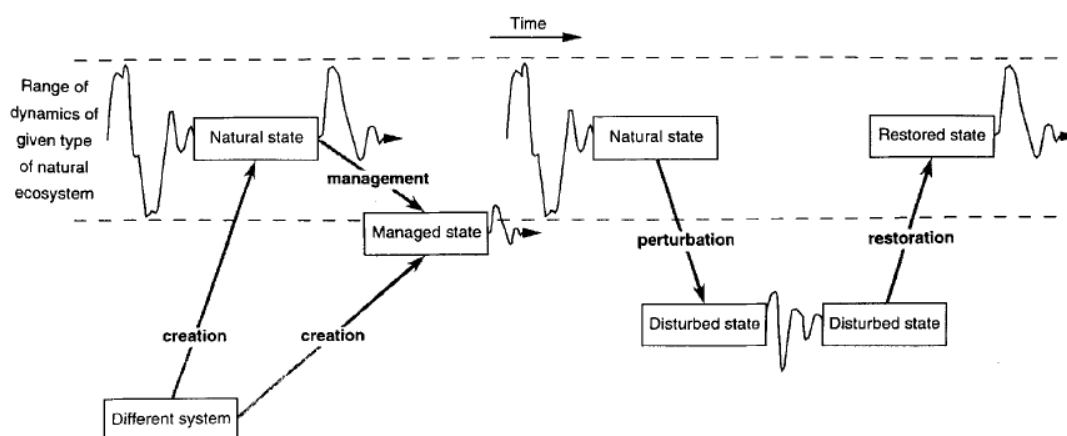


Figure 1 - Natural system and man-affected system dynamic through time (Henry & Amoros 1995).

Riverine restoration success depends on the knowledge in geomorphic, edaphic, hydrological, climatic and biological factors of the area to intervene. Indeed, the ideal situation would be that after the recovery of the hydrological regime and other river natural parameters the vegetation would return to its natural state. This process is usually denominated passive restoration

(Forget et al. 2013). However, in certain contexts (lack of propagules, need to prevent bank erosion) it is still needed a “little help” to start the regeneration process, and one of the most important decisions to make is the species to use for the rehabilitation project. A previous study, if possible, would help in these situations, in order to know the type of community present in the riverine corridor before the degradation. The autochthonous species to use, preferably, should be fast growing, well adapted to the riparian cycle of flood and dryness. Each restoration project defeats human impacts at multiple scales and specific levels of intensity and so each case must have a unique restoration/rehabilitation technique, in terms of species to be used, their ecological preferences as to water supply, how to plant them including spatial distribution and others (Melorose et al. 2009).

In Portugal there have been a few attempts to provide specific guidelines for river corridor restoration and/or rehabilitation. RIPLANTE project (<http://riplante.apambiente.pt/riplante/>) presents information on current habitat of the most common riparian tree species, however is still needed information on ecological requirements of trees at local scale, which is fundamental for restoration project implementation.

In the particular case of Odelouca river, associated with ecological mitigation of Odelouca dam construction, a previous study of Odelouca degradation state has been made (Fernandes et al. 2007). In this work along the degradation classification of the streams several restoration and rehabilitation recommendations are presented, mostly in the stream reaches more similar to the area being submerged by the dam. The restoration of riparian vegetation and margin and habitat rehabilitation are among the measures to be applied.

Another study proposed the rehabilitation of Odelouca through several techniques of bioengineering (Jesus 2008). The study of Jesus (2008) focus mainly on observation of environmental conditions present in the time of the field visits, in 2008, and trying to replicate those and even improve the fish cover in river by building new structures. In 2010, after the dam construction a rehabilitation program began in the framework of RICOVER project (Mendes et al. 2010). Vegetation dynamic was approached in a few case studies in Odelouca (Jesus 2008), Valverde and Pêra Manca (Colaço 1997; Pimentel 1999) rivers without many results that would be effective and important for restoration or rehabilitation of the river.

There is still research being developed in order to understand the best way to successfully establish tree species in riverine landscapes for riverine restoration. For example, Pimentel 1999 showed that *Fraxinus angustifolia* has good survival rates in a riverine restoration project in rivers Valverde and Pêra Manca, in Évora. However, the main problems with establishing the species were margin morphology, water availability in the soil and effects of water shortage during dry season. The first dry season after plantation is the most important for future establishment of tree species, water availability in the soil being the critical factor for survival. After establishment of the surviving individuals there are few (Colaço 1997; Pimentel 1999) studies following the growth of riparian species, namely *Fraxinus angustifolia*, in restoration projects as in natural systems.



With more knowledge of local conditions, like climate and water availability and their effect on tree growth, higher restoration/rehabilitation success rates could be achieved in future projects.

## 2.2 Dendrochronology

The study of the chronological sequence of annual growth rings in trees is called dendrochronology (Stokes and Smiley 1996). This science is based on the principle that a tree-ring width is determined by several variables (1): climate, age related growth trend, endogenous and exogenous disturbances pulses originating from within and outside, respectively, from the forest community, each with a higher or lower impact on the final width of each ring (Cook 1985).

$$(1) G = C + A + D1 + D2 + E \text{ (Cook 1985)}$$

Where G the well-dated tree-ring widths measured along a single radius, C the climatically-related growth variations common to a stand of trees including the mean persistence of these variations due to a physiological preconditioning and interaction and climate with site factors, A the age related growth trend, D1 the endogenous disturbance pulse originating from forces within the forest community, D2 the exogenous disturbance pulse originating from forces outside the forest community and E the series of more or less random variations representing growth influencing factors unique to each tree or radius within the tree.

The endogenous disturbances are easily confounded with the exogenous, making it not a very simple distinction. Endogenous disturbances are caused by factors of vegetation independent of the environment, like the normal cycle of growth and senescence, dominant and dominated trees, but these interactions in vegetation can be influenced by the exogenous disturbances like availability in water and nutrients. In Fritts (1976) limiting conditions are classified as two types, internal and external, which can be in some ways related to the endogenous and exogenous disturbances pulse, respectively. External limiting factors can be water, temperature, light, carbon dioxide, oxygen and soil minerals. Internal factors are amounts of available nutrients, minerals, growth regulators, enzymes and water.

Since early the main application of dendrochronology was tree dating, namely the use of tree rings to date events. This dating can be associated with other fields of knowledge like climatology, hydrology or ecology, and thus creating subfields of dendrochronology, such as dendroclimatology, dendrohydrology and dendroecology.

Trees have two types of growth: apical growth (increasing in height) and radial growth (increasing in width). Depending on the tree species growth environment there are very particular ways of annual ring production, for example, in tropical forests tree growth is continuous, the existent rings are attributed mainly to endogenous rhythms (Cook and Kairiukstis 1989). In temperate climates each year a layer of earlywood and latewood is formed

and these two types of wood create the tree-ring (figure 3). Earlywood is produced in the beginning of each growing season and latewood in the end, therefore the earlywood and latewood ring width depends greatly on environmental conditions during the growing season.

The earlywood of hardwoods is distinguishable by having vessels with bigger diameters and lighter color than latewood, that appears darker and with smaller vessels. When this pattern is clearly visible the wood gives the name of ring-porous species, like *Fraxinus angustifolia*. Species with pores evenly distributed, and not clearly distinguishable rings, are called diffuse-porous species, like *Alnus glutinosa*. This difference is more or less visible depending on the years and the tree species (figure 2).

One of the reasons that makes trees so important for dating hydrological or climate events is that it is possible to associate, with a high degree of accuracy, a calendar year to a single tree-ring, which allows to infer past climatic or hydrologic information from tree rings. Most of the dendroclimatic studies have been conducted in arid and semiarid areas, and in alpine ecotones, where there are strong limiting factors and thus the response of tree rings is very sensitive. In the last decades, several dendrochronological studies have been published using tree species from humid zones with complex eco-climatic factors and interactions, and it is frequent to find “complacent” tree-ring width series. Nowadays, the value of riparian species for dendrohydrological studies is widely accepted despite its limitations (Rodríguez-González et al. 2014).

For example, Mediterranean riverine species are even harder to analyze because their life length depends on the frequency of floods difficulting the cross-dating of short tree-ring width and so diminishing the certainty of the analysis with climatic and hydrological factors. Additionally, dating tree-ring width series from riparian species is also difficult, due to asymmetrical growth, presence of scars, intra-annual density fluctuations and missing rings (Ballesteros et al. 2010).



Figure 2- Sample of *Fraxinus angustifolia* with visible growth rings (photo by Inês Marques).

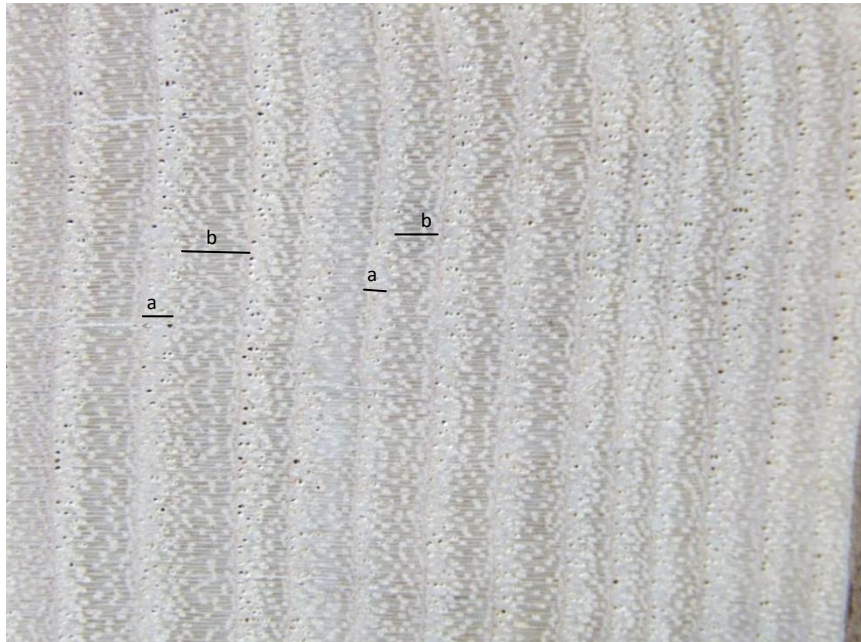


Figure 3 - Earlywood (a) and Latewood (b) in a sample of *Fraxinus angustifolia* (photo by Inês Marques).

### 2.3 *Fraxinus angustifolia*

The genus *Fraxinus* is distributed along northern hemisphere with *Fraxinus angustifolia* represented in southern Europe, north Africa and western Asia (figure 4), mainly around the Mediterranean basin (FRAXIGEN 2005). Its wood in Portugal is mostly used in river system rehabilitation and restoration.

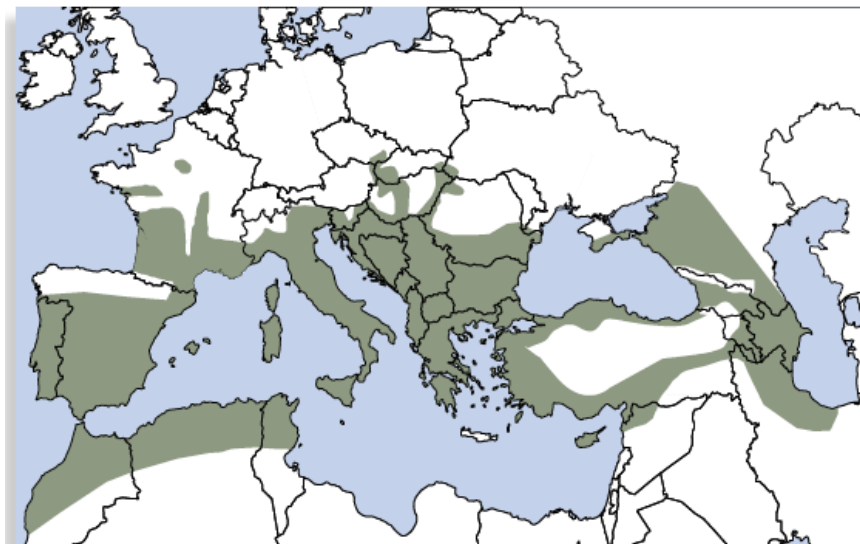


Figure 4 - Natural distribution of *Fraxinus angustifolia* in Europe. Source: FRAXIGEN (2005).

*Fraxinus angustifolia* (figures 5 and 6), commonly known as the narrow-leaved ash is a tree that grows near water courses, planes and floodable valleys and deciduous forests, normally where the groundwater level is deeper. *Fraxinus angustifolia* woods and riparian formations on intermittent Mediterranean water courses (with several species, including narrow-leaved ash) are part of the Habitats Directive 92/43/CEE (European Communities 1992) , adopted in 1992

for the conservation of natural habitats and of wild flora and fauna. These forests are designated as habitats 91B0, under habitat 91 Temperate Europe Forests, in the Annex 1 which describes all types of natural habitats of community interest whose conservation requires the designation of Special Zones of Protection (Comissão das Comunidades Europeias 2004).



Fig. 5 and 6 – *Fraxinus angustifolia* tree pending and on the right the detail of the leaf (photos by Patricia Rodriguez González).

In Portugal it is considered an indigenous species and is present in almost all the territory. Although the wood is appreciated for furniture and the leaves are used for cattle (ICNF 2013) there are not many commercial value in the market for this wood. Because it is an indigenous species by studying it we may have a better understanding of the type of growth and/or survival mechanism riparian trees use in Mediterranean streams to their particular climate, geomorphic and hydrological features.

*Fraxinus angustifolia* is a species present in Odelouca river, our study area, with very variable distances and heights to the river channel and less dependent on water availability as other riverine species like *Alnus glutinosa* (Rodríguez-González et al. 2014) also present in Odelouca and it prefers deep soils. In Odelouca basin it occurs in riparian formations associated with long or short periods of flooding, with species like *A. glutinosa*, *Salix salviifolia*, *Tamarix africana*, often in contact with upland species such as *Quercus suber* and *Quercus rotundifolia* and is a post-pioneer species (Rivaes et al. 2013). This species habitat is wider in regards to stream distance than other riverine woody species which prefer habitats close to the river channel. It can be found from up to no distance to the streamflow to 80m away from the stream.

Narrow-leaved ash is a fast growth species (Alameda and Villar 2012), which is important in cases of riverine restoration given the urgency to secure the seedlings in the river margins so that they will not be washed away by the annual periodic floods. Seedlings are more sensitive to soil physical properties than to chemical ones (Alameda and Villar 2012), which has still not

been entirely proven for the years proceeding seeds establishment. *Fraxinus angustifolia* seedlings are also known for their high tolerance to waterlogging, when compared with other riverine species and similar species like *Fraxinus excelsior* (Jaeger et al. 2009).

Root system and root uptake strategy is an important feature of riparian trees because even though near water courses there is higher water availability, tree species need to develop a root strategy that allows them to explore water soil, especially in periods of low or no flow. Common Ash (*Fraxinus excelsior*) roots compared with other riparian trees (like *Populus alba* and *Quercus robur*) explore less soil, therefore less density, having a sparse lateral extension with horizontal, vertical and oblique roots (Sánchez-Pérez et al. 2008). The water is obtained from non-saturated zones in contrast to other riparian trees like *Salix* spp, *Populus* spp (Sánchez-Pérez et al. 2008) and *Alnus glutinosa* (Rodríguez-González et al. 2014) which makes the phreatic water unavailable for common ash, except for trees in lower floodplain levels (Singer et al. 2014). In Singer et al. (2012) *F. excelsior* rooting depth was limited to the depth of fine sediment, not being able to penetrate gravel substrate.

## **2.4 Effects of environmental factors on ring width of tree species**

In this study climatic (temperature and precipitation), hydrologic (discharge), chemical (soil nutrient availability) and geomorphological (physical soil properties) variables were related to annual growth in *Fraxinus angustifolia* Vahl, as they are considered external factors to tree growth. Not much has been studied of the impact of these factors on riparian vegetation, especially on the narrow-leaved ash, mostly because of its geographic distribution restricted to the Mediterranean basin and lack of economic use. The information available is mainly about catastrophic events like flashfloods (Ballesteros et al. 2010).

When catastrophic climatic event happens along a drought the width of the ring formed that year will be narrower than the adjacent tree rings (Gasith and Resh 1999). However it still has not been shown a consistent pattern in earlywood-vessel formation for ring-porous species when responding to environmental factors (Sass-Klaassen et al. 2011). This shows that in order to understand if there are any factors limiting or enhancing growth or other anatomical features the responses of vegetation should be analyzed by species.

In a study in Central Europe (Lévesque et al. 2016) warmer temperature during vegetation period significantly enhanced the growth of *Fagus sylvatica* L., *Quercus robur* L. or *Quercus petraea* (Matt.) Liebl. and *Fraxinus excelsior* L.. Low nutrient availability and water deficits reduce the growth of *F. sylvatica*, *Abies alba* Mill. , *F. excelsior* and *Picea abies* (L.) H. Karst., explained because the species have their growth optimum in rich soils where water availability is not a limiting factor and not in excess.



Tardif and Bergeron (1993) found that *Fraxinus nigra* had a strong relationship between tree-ring width and weather conditions in the year before and the year of growth which was influenced by small variations in topography (under 1m). With *Fraxinus excelsior* in short periods of time (10 years) growth in stable or aggraded reaches is not significantly different when comparing different tree distances to the active channel, however there was a higher growth in plots less separated from the water table (Dufour & Piégay 2008). In period of low water availability *F. excelsior* can maintain moderate growth showing that it has resistance to water scarcity (Singer et al. 2012).

Although there are some responses as to the effects of climate and hydrological factors on *Fraxinus spp* growth the same is not true for soil properties like nutrients and granulometry. The lateral exchange of water, sediment and nutrients between river channels and their flood plains is an important and complex ecosystem process. The flood pulse concept of Junk et al. (1989) highlights the importance of these transfers for ecosystem functioning and the integrity of flood-plain river systems. Nutrients play an important role in regulating primary productivity in flood-plain systems because nutrients can be transferred to flood plains in association with sediments during overbank flows (Pinay et al. 1992). Soil nutrient availability is important not as a single factor for growth but has a complementary factor to catastrophic situations such as flood or drought in the years after and during these catastrophes. Drought and floods (or situations when plants roots are submerged by a certain period of time) both decrease nutrient concentration and uptake, but not all nutrients are affected the same way (Kreuzwieser and Gessler 2010).

Phosphorous (P) is one of the most important and limiting nutrients for life, mostly because it is present in the soil in very small quantities and not easily accessible, for it is mostly available for plant capture in the form of soil soluble inorganic P. Drought diminishes microbial activity which is the main responsible for mineralization in the soil, and therefore the transformation of nutrients to forms that can be used by plants. Several studies have shown that in drought situations the ratio between soil soluble inorganic P and soil soluble organic P decreases, therefore in drought periods plants have less availability to P and K in the soil (Sardans and Peñuelas 2004; Sardans and Peñuelas 2007). In artificially flooding it has been demonstrated that P availability increases with a rapid decrease following drainage (Wright et al. 2001), which due to flood-pulse concept might not have such a result in natural riverine systems because of the additional transport of nutrients between river and floodplains (Junk et al. 1989). The reasons for the increase in availability are still not totally clear. In environments with human addition of phosphorous to the system through fertilizers the runoff of the flood can add P to the systems downstream. The behavior of phosphorus is quite known in a drought or flood situation, but it has not been studied its direct relation with tree growth, and its effects on different species from several environments, that have particular needs and thresholds.

Nitrate ( $\text{NO}_3$ ) was significantly related with tree growth variables in *Pinus uncinata* (Sheppard et al. 2001) and it has been shown that there exists important interactions between water

availability and soil nutrients which are significant to tree growth responses to climate in *Fagus sylvatica*, *Quercus spp*, *Fraxinus excelsior*, *Abies alba*, *Picea abies* and *Pinus sylvestris* like Carbon and Nitrogen ratio (Lévesque et al. 2016).

The species *Fraxinus angustifolia* has been studied by Ballesteros et al. (2010) to assess the impacts of flash floods on the wood, which determined that there was a raise in vessel size and quantity caused by the flash flood. When studying river flow and precipitation the responses for narrow-leaved ash come alongside *Ulmus minor* in a study comparing native riverine species with invasive species (González-Muñoz et al. 2015) where it presented higher Basal Area Increment (BAI) with increase in spring river flow and spring precipitation, as well as summer river flow .

### 3 MATERIALS AND METHODS

#### 3.1 Study area

Odelouca river (figure 7) belongs to river Arade Basin, located in Southern Portugal, Algarve region, in a transition zone between Barrocal and Barlavento Algarvio. The river source is located in Caldeirão Mountain and it currently has a total length of 35.71 km (after dam construction in 2010), being considered as a natural stream by the Environmental Portuguese Agency (APA 2015). By the time the samples used for this study were collected (2009) the stream had a length of 92.6 km, maximum altitude of 460m and minimum of 1m with a medium slope of 0.5% (Ex-MAOTDR 2004). It is situated in two Special Protection Zones (Monchique and Caldeirão), with the code of PTC0037 and PTC0057 (respectively) of Nature Network 2000 (European Communities 1992).

Odelouca drainage basin is influenced by two mountains with high rainfall per year values in the Algarve region, those being Caldeirão and Monchique mountains with 1612mm and 2081mm per year (Ex-MAOTDR 2004) respectively, on top of the mountain. In average in all Algarve yearly rainfall values vary between 1277mm and 406mm, with about 80% of this rainfall happening in the humid semester.



Figure 7 - Spatial representation of the study area. Sources: <https://hyperscola.wordpress.com/ciencias-da-natureza/portugal/rios-de-portugal/> and Google Earth.



### 3.1.2 Geology and Climate

Even though having a mountain west of the stream the main climatic influence is Mediterranean and so it has a marked cold and wet and dry and hot season (ARH and APA 2012). According to Köppen classification the hydrographical region of Algarve streams has a climate type Csa, temperate mesothermic with a wet and dry winter (Cs) and dry summer (a) (ARH and APA 2012). According to Thornthwaite classification the region is sub-humid dry, particularly in Monchique it is dry (ARH and APA 2012).

Therefore this region is very likely to be affected by floods in the winter/autumn and droughts in the summer. It has been recorded a total of 5 important droughts and floods since the 1970's in Algarve region. From 1974 to 1976 all Algarve region experienced a two year drought (Ó and Monteiro 2005) and three droughts in Algarve streams in the periods of 1980-1983, 1990-1994 and 2002-2006 (Vivas and Maia 2007). These droughts are considered the most severe and dry of the last 50 years, according to Standardized Precipitation Index (SPI) and Normal Precipitation (NP). Only one flood has been registered in the streams of Monchique and Odelouca in October of 1997 (Lorena 1998), in Monchique its discharge and precipitation with a 1000 year return period and in Odelouca with a return period of 10 to 20 years.

The Monchique mountain plays an important role in the geomorphological properties of the area, it stands between the plane area of Alentejo and Algarve. The area of Algarve Mountain is mainly lithological homogenous, composed of alternated layers of schist and greywacke and Barrocal is formed by Mesozoic carbonated rocks (ARH and APA 2012).

In terms of territorial management Odelouca river belongs to the so called group Streams of Algarve (Ribeiras do Algarve), for which management and territorial plans are made every 4 years, the most recent of June 2015 (<http://www.apambiente.pt>). Nowadays Odelouca has been divided in three administrative regions: one belonging to the dam basin and the other two to the natural stream.

The hydrographic region has a total of 23 underground waters with areas ranging from 5 to 300 km<sup>2</sup>. The area is mostly occupied by natural and semi-natural forests (36.1%), water bodies (30.9%) and agriculture and agro-forestry (27.6%). The hydrographical region of Algarve Streams is very rich in underground water resources (ARH and APA 2012).

Benafátima stream joins Odelouca river on its right margin and is an important feature for this project. It is the biggest stream that runs to the studied sector of Odelouca river, and according to previous studies a major parameter in river ecology, providing environmental conditions that permit to sustain certain types of vegetation more exigent in water availability like *Alnus glutinosa* (Rodríguez-González et al. 2014).

## 3.2 Climatic and Hydrological Data

A total of 8 independent variables were used as possible climatic and hydrological factors affecting tree growth of *Fraxinus angustifolia*. Those variables were: daily and annual discharge (in m<sup>3</sup>/s); maximum and minimum temperature (in °C); mean annual precipitation (in mm), daily and annual streampower and discharge variation for designated year periods (in m<sup>3</sup>/s).

### 3.2.1 Precipitation and Temperature

Monthly climatic data (precipitation, maximum and minimum temperature) were obtained from the nearest 0.5°x0.5° grid point for the last 100 years (<http://climexp.knmi.nl/>). As opposed to drainage area and discharge, the climatic variables were not representative of each tree individually, but of the Odelouca basin. These climatic variables were used in the primary statistical analyses with Pearson's correlations.

### 3.2.2. Drainage Area and Discharge

In order to obtain the discharge value to each individual tree, it is essential to take into account the precipitation on each basin area for each tree during the period of time that we want to analyze for discharge and the basin drainage area. Precipitation values for each month of the year since 1967 were obtained (from [www.SNIRH.pt](http://www.SNIRH.pt)) for the 9 precipitation stations closer to Odelouca river. Precipitation on the river drainage basin for each tree was interpolated from Thiessen Polygons, using Arctoolbox in ArcMap version 10.0, from these 9 precipitation gauge stations (table1).

Table 1 - Station information of Code, Name, Altitude (m), Latitude, Longitude and Basin

Station Code	Name	Type	Altitude (m)	Latitude (°N)	Longitude (°W)	Basin
30G/01UG	Alferce	Pluviometric	325	37.33327	-8.49058	Arade
30G/03C	Barragem do Arade	Pluviometric	58	37.23800	-8.37500	Arade
30H/01U	Foz do Ribeiro	Pluviometric	142	37.31540	-8.23466	Arade
30F/01C	Monchique	Pluviometric	792	37.32278	-8.59456	Arade
30I/01U	Monte Ruivo	Pluviometric	269	37.28207	-8.15132	Arade
30H/04UG	Santa Margarida	Pluviometric	246	37.24700	-8.19100	Arade
29I/01UG	São Barnabé	Pluviometric	259	37.35907	-8.16349	Arade
29G/02G	São Marcos da Serra	Pluviometric	130	37.36667	-8.38098	Arade
30H/02U	Vale de Barriga	Pluviometric	92	37.28206	-8.30133	Arade
30G/01H	Monte dos Pachecos	Hydrometric	55	37.30000	-8.46670	Arade

Mean annual discharge values for each tree were calculated (equation 2) using data of annual discharge from Monte dos Pachecos station, precipitation on the river drainage basin for each tree, precipitation on the river drainage basin for Monte dos Pachecos (both interpolated from precipitation values from 9 different stations), drainage areas for each tree and Monte dos Pachecos (figure 8).

$$(2) Q_a = Q_k \times \frac{A_a}{A_k} \times \frac{P_a}{P_k}$$

For this equation, read:  $Q_a$  is Modular discharge for each tree ( $\text{m}^3/\text{s}$ );  $Q_k$  is annual discharge for Odelouca basin obtained from gauge station Monte dos Pachecos ( $\text{m}^3/\text{s}$ );  $A_a$ = drainage area for each tree ( $\text{km}^2$ );  $A_k$ = drainage area for Monte dos Pachecos ( $\text{km}^2$ );  $P_a$ = calculated annual precipitation on the river drainage basin for each tree (mm);  $P_k$ =precipitation on the river drainage basin for Monte dos Pachecos (mm).

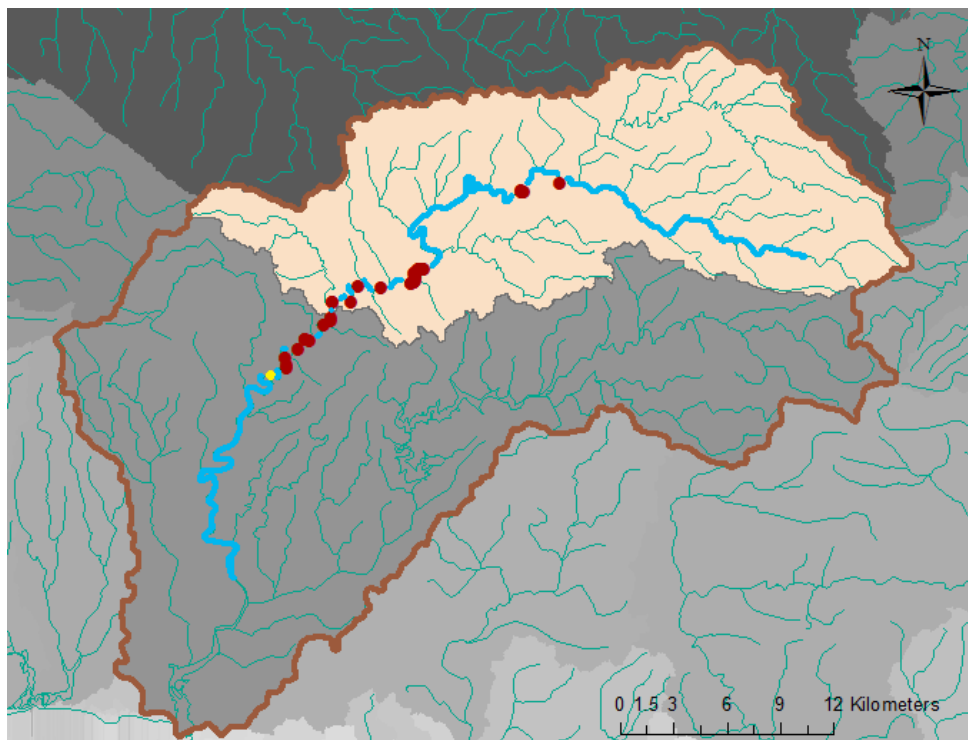


Figure 8 – Spatial representation of Arade basin area, Odelouca river, sampled trees and Monte dos Pachecos station. Arade basin is represented with contour in brown and example for drainage area for one tree in pink. Red dots representing sampled trees, yellow dot representing Monte dos Pachecos hydrometric station and Odelouca river highlighted in blue.

### 3.3 Soil Sample collection

#### 3.3.1 Soil collection

Every sampled tree (described below) had a correspondent soil sample taken a composite sample from the topsoil up to a depth of 30-40cm. When trees were close from each other and

the soil maintained the same visible appearance, only one soil sample was made for the group of trees. In total 37 soil samples were collected.

### **3.3.2 Soil analysis**

There were a total of 28 soil samples analysed, less than the number of trees because when several trees were close to each other and considered to be in the same type of soil, only one sample was selected for analyses.

#### **3.3.2.1 Physical Analysis**

Each sample was dried after field collection and then divided in half, each with a minimum weight of 250g. One part was used to access the soil structure (constructed by the particles and conglomerates, as well as non-mineral elements) and the other part was used for chemical characterization (pH,  $\text{NO}_3$ ,  $\text{NH}_4$ , Conductivity,  $\text{P}_2\text{O}_5$  and  $\text{K}_2\text{O}$ ).

Soil structure was examined by the proportion of particles or aggregated of particles diameter: <0.05mm (clay and silt), 0.05-0.1mm (very fine sand); 0.1-0.25mm (fine sand); 0.25-0.5mm (medium sand); 0.5-1mm (coarse sand); 1-2mm (very coarse sand); 2-5mm; 5-8mm; 8-10mm and >10mm, a total of 10 categories. Sand diameters below 2 mm were categorized according to USDA.

Each sample was mechanically sieved during 15 minutes and after each soil category obtained was weighted in a scale with an estimated error of 0.001. Each proportion of soil category was calculated simply by dividing each weighted category by the total weight of the sample, before sieving. This allowed calculating the amount of soil lost during the procedure, none of the samples had lost more than 5% with the mean percentage of lost soil of 0% and so, every soil sample was used for further analysis.

#### **3.3.2.2 Chemical Analysis**

In order to understand the effect of nutrients and other chemical components on tree growth each sample was analyzed for the most important nutrients in tree survival: P, K, N and C. Due to previous results in nutrients effect on tree growth (Sheppard et al. 2001; Lévesque et al. 2016) from each sample was extracted ammoniacal nitrogen ( $\text{NH}_4$ ), nitric nitrogen ( $\text{NO}_3$ ), phosphorus pentoxide ( $\text{P}_2\text{O}_5$ ) and potassium oxide ( $\text{K}_2\text{O}$ ). The phosphorus pentoxide and potassium oxide was obtained by the EGNER- RIEHM method and for the two nitrogen compounds an extraction with KCL 2M, readings in the self-analyzer of segmented flux and colorimetry were made.

The percentage of Organic Matter (O.M) was obtained by dry combustion to 1200°C by infrared light.

In addition to the 28 soil samples collected in 2009, two more soil analysis of C/N and Organic Matter were available from a previous study in the same river (Rivaes et al. 2013), so they were used for the latter analysis, together with the other samples.

### 3.4 Wood Preparation and Tree-Ring Identification

#### 3.4.1 Wood Sampling

Initially a total of 70 *F. angustifolia* trees were sampled along approximately a 37 km reach of Odelouca river (figure 9). When sampling 13 trees showed partially rotten stems and for that reason were discarded. The code for core identification had a total of 8 characters and it had information about the site where it was collected, the species, number of the individual tree and which core. As an example, a core from the tree 008 had a code like this: odfa008a, “od” means that it comes from the Odelouca river, “fa” means that its species is *F. angustifolia*, 008 is the number of the tree – normally corresponded to the order of sample collection- and “a” designs the radius, that could go up to letter “d”, which means four radii.

For each tree, individual parameters were collected (field sheet in annex 1): species, height breast diameter, height, number of stems, slope, latitude, longitude, distance and height to river, soil sample, presence of fire in the area, trashline height, diseases, erosion and type of river habitat. Any visual parameters showed at site that could be interesting or help further analysis were annotated.

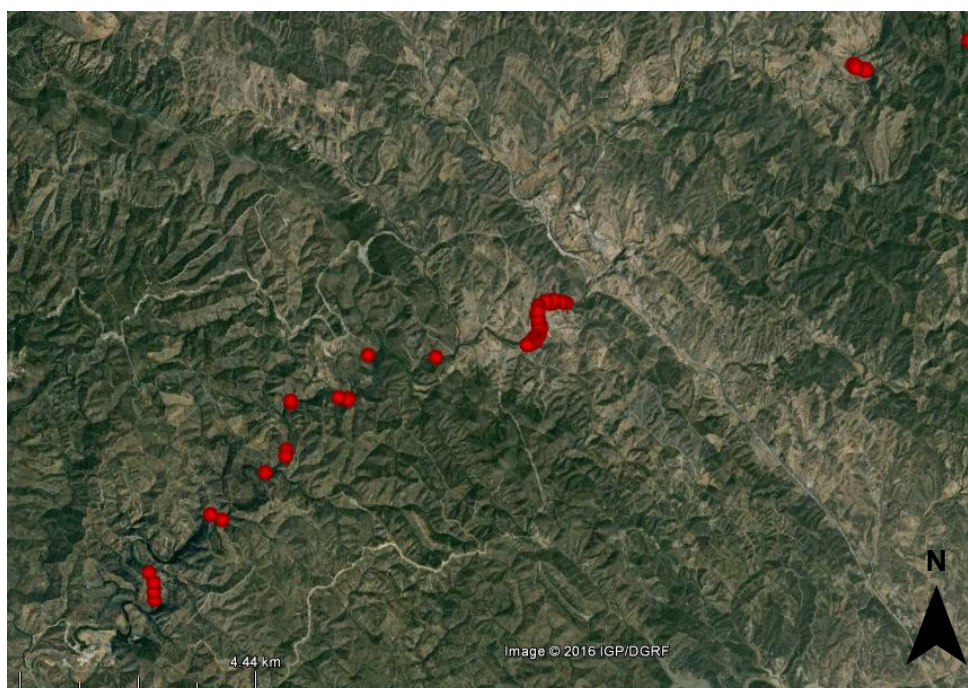


Figure 9 - Sampled trees in Odelouca river. Source: Google Earth.

### 3.4.2 Sample preparation

A total of 53 trees of *F. angustifolia* were sampled (28 increment cores and 25 tree sections) with at least 2 radii at breast height during summer of 2009. When possible tree sections were retrieved, and brought for analysis alongside the increment cores (figures 10 and 11). After sampling, cores and cross-sections were dried in a natural environment, the cores were later glued to a wooden frame. After dried each disk surface underwent a planing treatment before sanding and cut in pieces to facilitate manipulation so that future analysis would be possible.



Figures 10 and 11 – Increment cores (top) and tree sections of *Fraxinus angustifolia* after sanding (photos by Inês Marques).

In order for the rings to be visible for measuring, the samples were first sanded with paper grains 100 to 180. After this there was a primary ring analysis through a magnifier, in order to identify rings and other anomalies present in the wood like missing rings, false rings and intra-annual variations. These anomalies are important in order to make the correct correspondence between tree rings and year. In each tree every time it was possible at least three rays were identified for later ring-width measurement.

### **3.4.3 Visual Analysis and Ring Identification**

Tree rings were measured with an accuracy of 0.01 mm on a LINTAB digital positioning table connected to the software TSAP (Rinntech, Heidelberg, Germany). Individual tree-ring series were visually cross-dated and statistically verified using TSAP and *detrendedR* package (Campelo 2012) in R program version 3.2.0 (R Development Core Team 2008). Initially visual cross-dating was made and the worst series (that did not match with the mean) were excluded, followed by statistical confirmation, in which all radii with a correlation lower than 0.25 were discarded. Series with correlations higher were selected, making the final group of series used for this study a total 94 radii, representing 43 different individuals (11 individuals with just 1 radius).

## **3.5 Statistical Analysis**

Several approaches were used to analyze the ring-width data. In addition to analyze the total number of samples, two types of groups were made in which all chronology analysis was based on: group Far/Near and Downstream/Upstream. Analysis of BAI, cambial age, pointer-years were determined, simple correlations and generalized linear mixed models were applied using these same groups. All analyses were performed using *R* software, version 3.2.0 (R Development Core Team 2008).

### **3.5.1 Work groups**

In order to analyze if there were any responses on tree-ring growth depending on tree distance and height to the river channel and distance to source, trees were separated in groups showing the effect of transversal and longitudinal gradients in the river. Tree position along these gradients influence tree access to water, nutrients and their exposure to geomorphic disturbance during floods. For the group studying the relative transversal position to the river, trees with less or equal to 2m height from the river and 8m or less of distance from the river active channel belonged to the Near group and the Far group had all individuals with more than 2m height and 8m distance from the active river channel. The individuals that did not fall into this classification were later added to each group with the help of a Principal Components (PCA) (Wickham 2016) and field observations. After grouping all individual trees, group Far had 18 individuals (35 cores) and group Near had 25 individuals (59 cores).

The other two groups were made based on the tree position according to the longitudinal river profile in relation to the confluence of a large tributary (Benafátima stream) at approximately the middle of the river length of study. In total 16 individuals (43 cores) belonged to the Downstream group and 27 to the Upstream group (with 51 cores).

### **3.5.2 Growth patterns of population and work groups**

#### **3.5.2.1 Dendrochronology**

Chronologies were developed for both groups and the totality of trees, previously selected by visual and statistical analysis. The final chronologies were made with the *detrendeR* package (Campelo 2012), using a twostep detrending with a spline length of 30 years fitted to each core. Auto-regressive model was applied to each core to obtain a residual chronology. The Expressed Population Signal (EPS) was calculated in order to indicate how well the obtained chronology represented the signal of a perfect chronology (i.e an infinitely replicated chronology) (Wigley et al. 1984; Briffa and Jones 1990).

#### **3.5.2.2 Basal Area Increment**

The ring-width series were converted to ring-area series based on the distance between the innermost measured ring of the tree and the pith, this method assumes a circular cross section and the area of each ring called Basal Area Increment (BAI). Not every core had pith, and for those without pith the missing distance was estimated using the average of all series with pith, by age class. Three age classes were used: less than 40 years, between 40 and 80 years and more than 80 years. BAI was calculated using *dpIR* package (Bunn 2008; Bunn 2010; Bunn et al. 2015).

BAI was calculated for all trees and for the two categories in each of two work groups Far/ Near and Downstream/Upstream. The period used for this analysis was 1932-2009 to guarantee at least 5 values for each year.

#### **3.5.2.3 Cambial Age**

Using cambial instead of tree age made possible to analyze the effect of cambial age independently from that of calendar year. Samples showing pith were used to determine cambial age and to obtain a predicted growth curve. This curve was used to estimate the cambial age of radii without pith.

Then, all radii were aligned by its cambial age and smoothed with a 20 year spline curve, using *detrendeR* package (Campelo 2012). The obtained curves were used to compare Near *versus* Far and Downstream *versus* Upstream.



#### **3.5.2.4 Pointer years**

The comparison of extreme growth events (narrower and wider rings) with climatic data can be used to better understand the growth-limiting factors (Neuwirth et al. 2004). These extreme growth events (positive or negative) are given by pointer years. A negative (positive) pointer year is formed when several trees, growing in the same region, show a narrower (wider) ring in response to unfavorable conditions for tree growth (Schweingruber et al. 1990). Additionally other ring properties such as latewood/earlywood proportion, vessel lumen and intra-ring wood density can be used as ecological indicators of any environmental factor influencing tree growth.

Pointer years were calculated using the relative growth change method described by Schweingruber et al. (1990) with *R* package *pointRes* (van der Maaten-Theunissen et al. 2015), which relates tree growth in year *i* to the average growth of *n* preceding years. The parameters used for this analysis were: 6 preceding years, 50% threshold above which a relative growth change is considered a positive event year, 30% threshold below which a relative growth change is considered a negative event year and 70% as minimum percentage of trees that should display a positive (or negative) event year for that year to be considered as positive (or negative) pointer year (Maaten-Theunissen and Maaten 2016).

Pointer years within these parameters were calculated for all trees and groups Far, Near, Downstream and Upstream. The period used for this analysis was 1932-2009 to guarantee at least 5 values for each year.

### **3.5.3 Environmental factors and growth**

#### **3.5.3.1 Population response to climate and hydrology**

Simple Pearson Correlations between tree-ring width, monthly total precipitation and mean temperatures (minimum and maximum) and discharge were calculated for the period of 1967-2009 for all trees, 1973-2009 for Near group, 1974-2009 for Far group and 1979-2009 for Downstream and Upstream groups with a significance  $\alpha=0.05$ . The data used for temperature – mean, minimum and maximum- and precipitation had no missing values however discharge data had up to three years gap, those were left with no observations.

#### **3.5.3.2 Tree growth response to multiple factors**

A matrix containing all the information retrieved on the field sample, climatic data, hydrological data and ring growth data was assembled, making a total of 255 variables.

The first step included data exploration to detect possible problems in raw data, to explore relationships within and across explanatory and dependent variables and to aid in the selection

of the final explanatory variables set used for linear models (Zuur et al. 2010). Outliers presence was studied with boxplots and Cleveland dotplots with packages *car* (Fox and Weisberg 2011) and *Hmisc* (Frank et al. 2016). Next step was testing for homogeneity and normality of the explanatory variables like the growth values for each year and other periods chosen for climatic or hydrological reasons.

The homogeneity and normality assumptions were tested through histograms, Bartlett's test and Shappiro-Wilks test using package *stats* (R Core Team 2015). If the *p-value* was superior to 0.05 the variables were considered to have a normal distribution and homogeneity. The ones that were heterogeneous were discarded from further analysis and distributions for the non-normal variables were assumed using package *fitdistrplus* (Delignette-Muller and Dutang 2015). All the BAI variables had a skewed distribution and were log-transformed so that the dependent variable was  $\log(\text{BAI}+1)$ , protocol followed by similar study of Lévesque et al. (2016).

To test for collinearity between the covariates several statistical tools were used, including PCA using package *evaluate* (Wickham 2016), Variance Inflation Factor (VIF) using package *car* (Fox and Weisberg 2011) values and correlation plots with Pearson's correlation using package *ellipse* (Murdoch and Chow 2013) and *car* (Fox and Weisberg 2011).

In the PCA ordination space (Kassambara and Mundt 2016), variables pointing to very close or totally opposite direction were considered as too collinear and only one from those was selected.

Pearson's Correlations were made with critical values for analysis of 0.5 and 0.8 and VIF values were attributed to each variable. If a variable had a VIF superior to 10 it would be discarded until all variables from the selected group had values below 10. To explore the relationship between the covariate and the response variables, bivariate scatterplots were used. Scatterplots enabled to visually assess relationships while keeping in mind that the lack of a clear two-way relationship does not mean that they have no association at all, there could still be more complex relationships involving more variables.

And so, having in mind all the different analysis in exploring the existent data a total of 4 independent and 6 dependent variables were chosen to be used in modelling.

Mixed models allow the understanding of what is the best independent variables combination that would explain tree-growth, with the addition of random effects on the model. Models were fitted at the tree level using packages *glmmADMB* (Fournier et al. 2012; Skaug et al. 2016) for variables with non-normal distribution (generalized linear mixed models) and *nlme* (Pinheiro et al. 2016) for normalized distribution variables (linear mixed models).

Stand was considered a random effect in linear mixed models for normal and non-normal distribution variables and for BAI variables the distance between the center of the tree to the year considered as study variable was calculated and trees gathered in three age classes, which were nested within the stand.

Each of the 4 independent variables were combined in all possible ways, between models with only random effect, one variable and multi-variables models, making a total of 16 tested models without interactions for each response variable. In order to find the best fitted model for the response variable, each was ordered by its AIC value and the likelihood of each model (or weight) was calculated using formula (3) and the probability of each model  $g_i$  ( $w_i$ ) was computed by formula 4 (Burnham et al. 2011). When sample sizes are small, like in this case and in most practical studies, a second order bias correction for AIC was calculated, called as AICc. AIC and AICc converge as sample size increases.

$$(3) \hat{A}_i = \mathcal{L}(g_i | \text{data}) = \exp(-(1/2)\Delta_i)$$

$$(4) w_i = \text{Prob} \{ \text{model } g_i | \text{data} \} = \hat{A}_i / \sum_{j=1}^R \hat{A}_j = \ell_j$$

Where:  $\hat{A}_i = \mathcal{L}(g_i | \text{data})$  = relative likelihood of model  $g_i$ ;  $\Delta$  = difference between AICc's of the model.

The best models were considered the ones with the highest weight and all models with a  $\Delta$  less than two with the best fitted model were considered alternative models; they must also have explanatory variables with a p-value inferior to 0.05. These models residuals were tested for normality and close to zero distribution in order to be accepted has statistically reliable and further valid discussion of the results.

## 4 RESULTS

### 4.1 Tree work groups

The final distribution of the data by groups (Near/Far and Downstream/Upstream) assigned using PCA (annex 5) and field annotations is presented in the annex 6. After grouping all individual trees, group Far had 18 individuals (35 cores), group Near had 25 individuals (59 cores), group Upstream had 27 individuals (51 cores) and group Downstream had 16 individuals (43 cores).

### 4.2 Climatic and Hydrological Conditions

#### 4.2.1 Temperature and Precipitation

The precipitation and temperature trends obtained from the exported data are visible in the figures below (figures 12, 13 and 14).

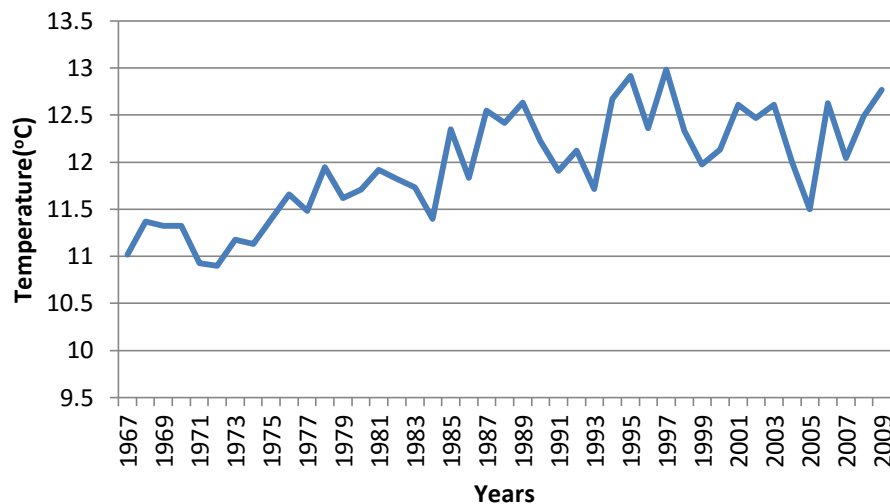


Figure 12 –Average monthly minimum temperature in Odelouca river per year since 1967. Source: <http://climexp.knmi.nl/>.

Mean minimum temperature ranged between 11°C and 13°C since 1967, showing an increasing trend for the last 40 years. Mean maximum temperature also shows the same trend, but with less amplitude and ranging from 19°C to 21.5°C, with the highest value in 1996 and the lowest in 1972.

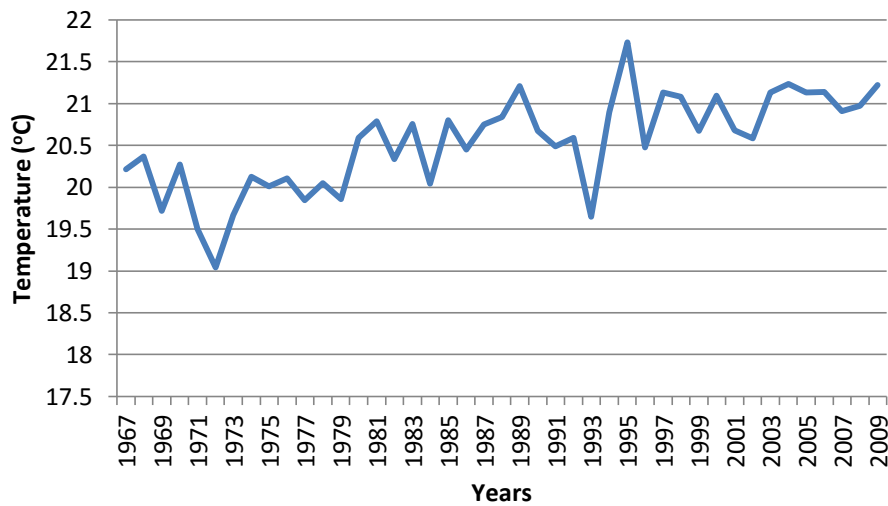


Figure 13 – Average monthly maximum temperature in Odelouca river per year since 1967. Source: <http://climexp.knmi.nl/>.

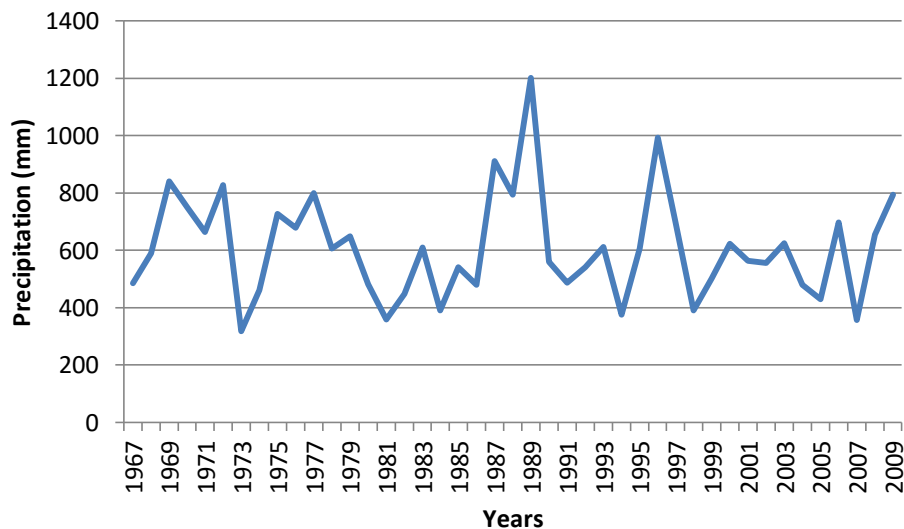


Figure 14 – Average monthly precipitation in Odelouca basin per year since 1967. Source: <http://climexp.knmi.nl/>.

Annual total precipitation (figure 14) ranged between 312mm, in 1973, and 1200mm, in 1989, maintaining stable fluctuation around 600 mm in the presented period. Years with higher values of precipitation are 1989 and 1997 which corresponds with a extreme flood in Odelouca river.

#### 4.2.2 Drainage Basin Area and Discharge

Mean annual discharge for all trees is presented below (figure 15), where it is very visible a low/near to zero annual discharge values in years 1971, 1982, 1983, 1984, 1991, 1992, 1993 and 1994. These values are consistent with historical drought periods of 1980-1983 and 1990-1994.

Higher values of discharge frequently occur in the years following the small discharge values, with a periodicity of around 10 years. The highest value since 1965 was in 1996, followed by 1997, the registered in Odelouca and Mochique rivers.

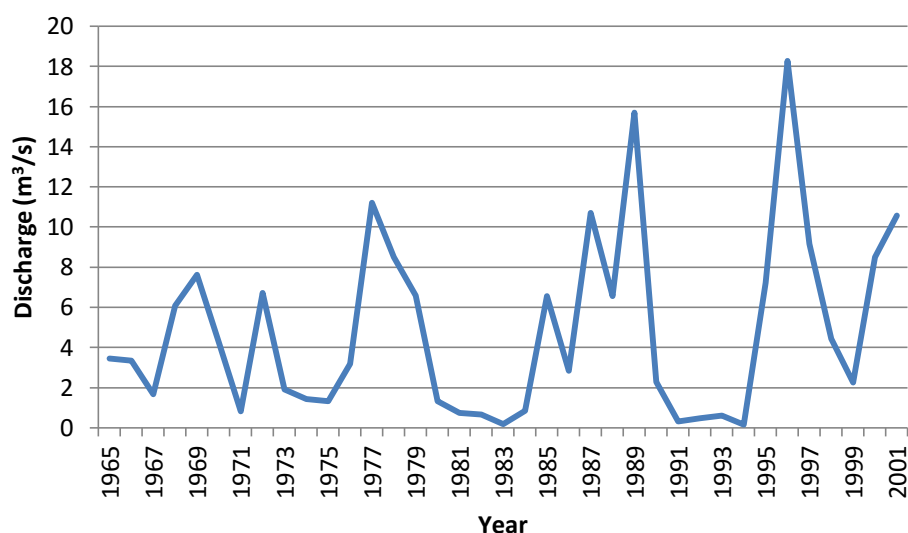


Figure 15 - Mean annual discharge in Arade basin per year since 1967 from Monte dos Pachecos station.

The closer to the source the smallest is the mean annual discharge value for each tree, varying from 0.97 to 2.55 m³/s (annex 7). The trees farther away from the Monchique mountain have a mean annual precipitation smaller than all the other trees, closer to Monchique.

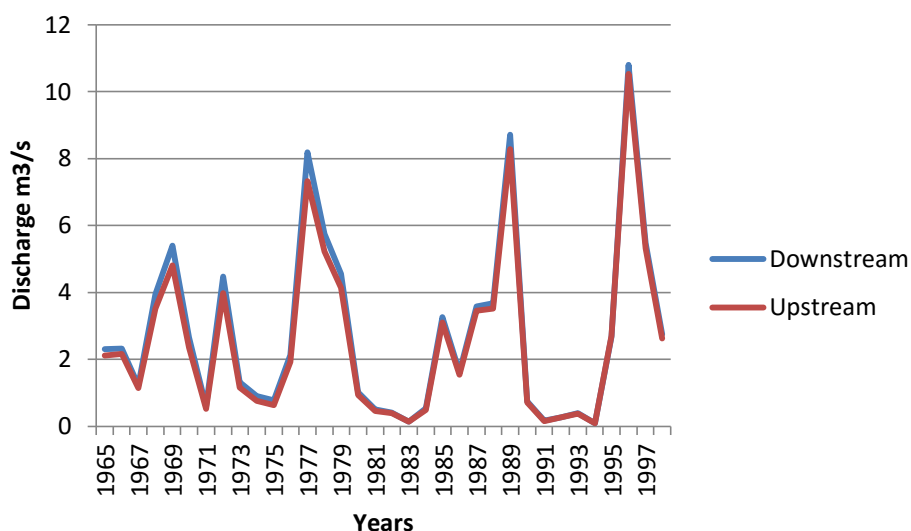


Figure 16 - Annual Discharge in m³/s from Downstream and Upstream subgroups.

Mean Annual Discharge for trees Upstream is lower than trees located Downstream (2.27 m³/s for the first and 3.3 m³/s for the second), a tendency also present when comparing discharge values along the years (figure 16). The discharge values are very similar since 1965, although

generally higher for Downstream group, with the most difference between the two groups in the years with the highest discharges.

### 4.3 Physical and Chemical Soil Properties

Table 2 - Soil chemical analysis results for all trees

	Units	Mean	Maximum	Minimum	n
<b>C</b>	%	1.70	2.78	0.67	31
<b>O.M</b>	%	2.92	4.80	1.15	31
<b>NH<sub>4</sub><sup>+</sup>-N</b>	mg/kg	8.87	17.47	5.42	28
<b>NH<sub>3</sub><sup>+</sup>-N</b>	mg/kg	1.08	6.80	0.01	28
<b>Total P<sup>a</sup></b>	ppm	10.91	53.52	0.16	28
<b>Total K<sup>a</sup></b>	ppm	66.29	124.00	39.00	28
<b>pH</b>	-	5.88	6.79	4.66	28
<b>Conductivity</b>	µs/cm	238.34	537.60	96.04	28
<b>C:N Ratio</b>	-	19.40	35.29	10.01	28

<sup>a</sup> Total P was measured as  $P_2O_5$ , total K as  $K_2O$

In general there were no high differences between the maximum and minimum values for each variable (table 2); the amplitude being within the normal scales. In regards to C, O.M, NH<sub>4</sub><sup>+</sup>-N, conductivity and C/N the values of mean, maximum and minimum are lower in Upstream (annex 4) compared to Downstream group. The same is observed between Near and Far groups (annex 4), but not with maximum values, which are the same in multiple variables.

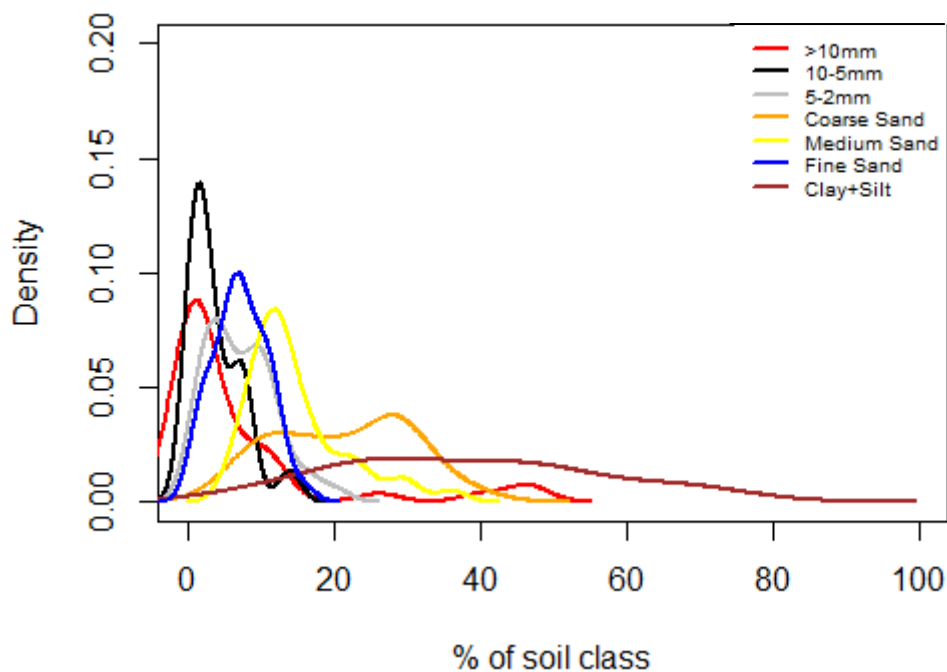


Figure 17 - Density of soil diameter categories for all sampled soil trees.

Most soil samples had high percentages of medium sand, coarse sand and clay and silt. With lower percentages but represented in more samples, there were bigger particles (10 to 5 mm)

and fine sand (figure 17). All samples were retrieved 30 to 40 cm below ground and so the distribution of the soil fraction might not coincide with what would be the expected for riverine soil.

Far and Near soil samples (figure 18) did not show much difference between their composition, both had high percentages of clay and silt on the samples. Most samples had low percentages of fine sand as well as particles from 5mm diameter forward.

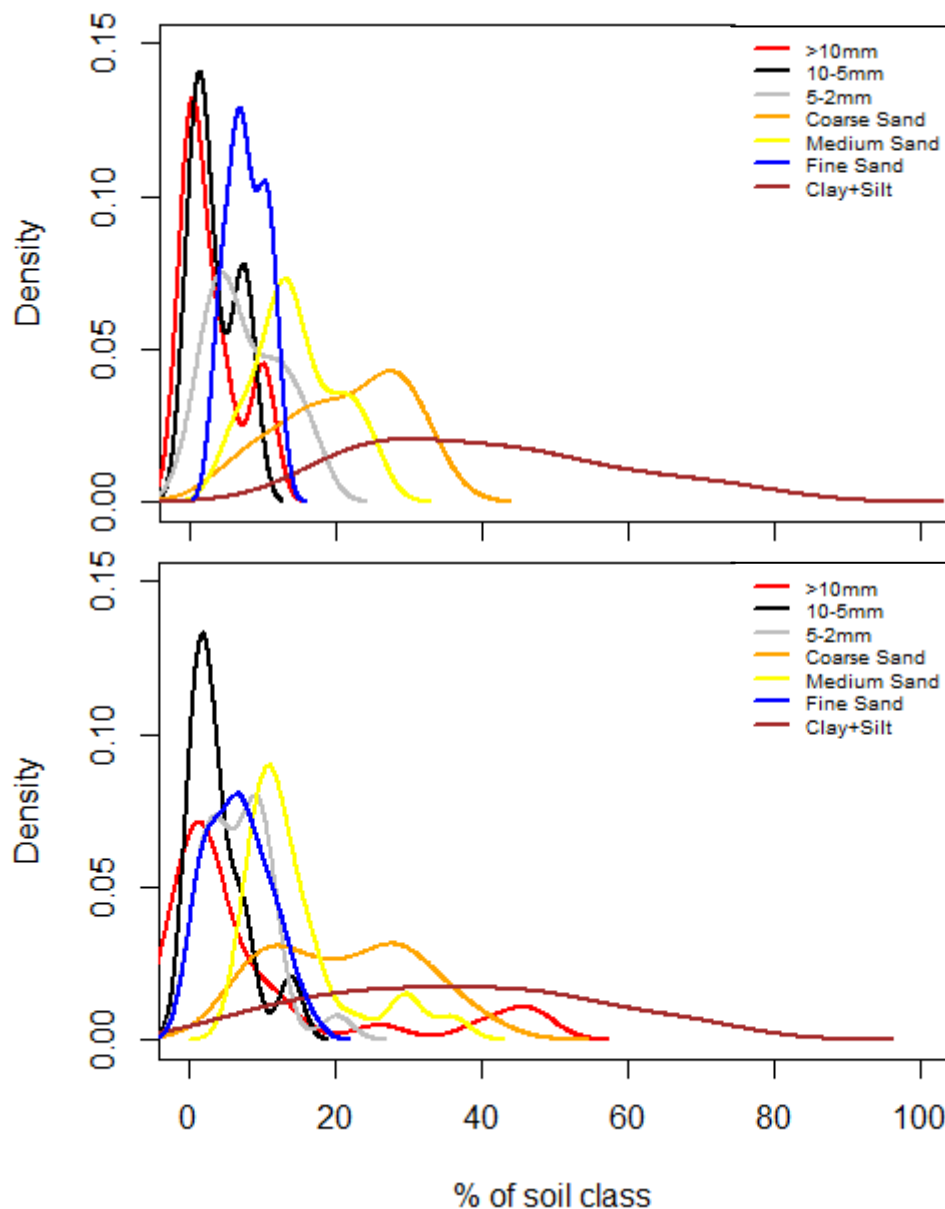


Figure 18– Density of soil diameter categories for sampled soil trees from Far (above) and Near (below) groups.

Downstream and Upstream groups were the ones with the most differences in soil portions constitution (figure 19). Downstream had higher portions of coarse and medium sand in the samples (both in percentages and density) compared to Upstream. That was expected because



water flow can transport farther away smaller particles, leaving the biggest and heaviest nearest to the river source.

The samples used for the physical and chemical analysis were collected in the summer of 2009 and since then have been stored at a room stable temperature environment. Therefore there is a probability that the fractions obtained in this analysis are not completely coincident with the ones experienced in natural state, because particle agglomerates can be very fragile and brake when not handled carefully. However, we can say with sure that some of the samples were still in their natural state because of the amount of boulders contained or the lack of agglomerates, which the good sense tells that either are samples with very high percentages in fractions above 2mm and low on the others, or with high percentages in fractions of fine and very fine sand and not in rocks.

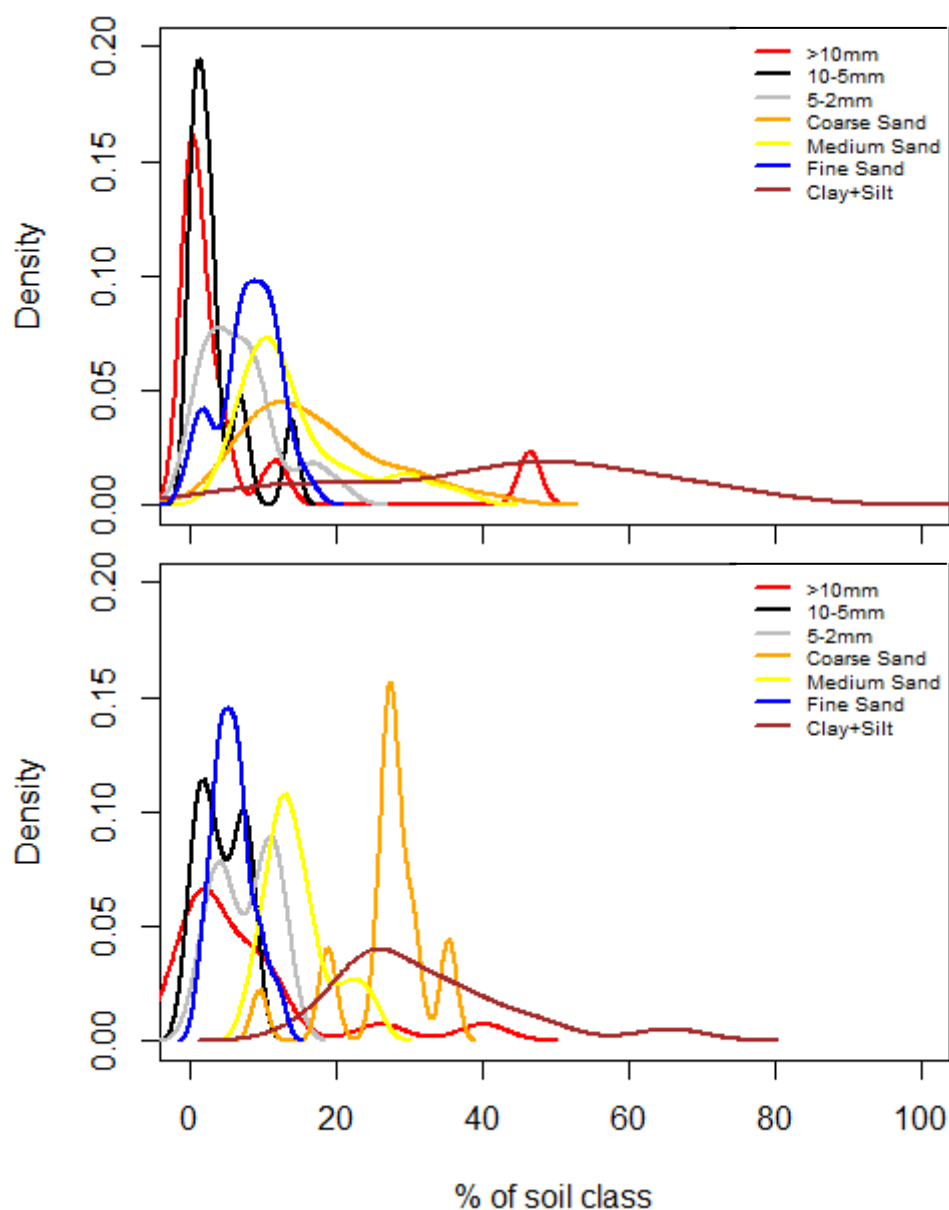


Figure 19 - Density of soil diameter categories for sampled soil trees from Upstream (above) and Downstream (below) groups.

#### 4.4 Growth patterns of population and work groups

In a preliminary analysis of raw tree-ring width (table 3) analyzes was made between groups according to position to active water channel (Far/Near) and to distance to source (Downstream/Upstream). The highest values of maximum and minimum raw tree-ring were in groups Far and Upstream, however the highest average tree-ring growth were in groups Far and Downstream (always comparing between groups Far/Near and Upstream/Downstream).

Table 3 - Annual tree-ring width (mm) information of average, minimum, maximum and mean span of raw data by groups

	All	Far	Near	Downstream	Upstream
<b>Average</b>	4.13	4.84	3.69	3.99	4.25
<b>Minimum</b>	1.79	1.81	1.79	1.79	1.81
<b>Maximum</b>	12.49	12.49	5.92	6.68	12.49
<b>Mean Span</b>	48.00	47.00	50.00	58.00	40.00

##### 4.4.1 Tree-Ring Width Chronologies

Tree-ring width chronologies were obtained from individual ring-width measurements series. There was an initial verification for any missing rings in the cores. More than 70% of all the series had more than 59 years, three trees had a length up to 110 years (figure 20).

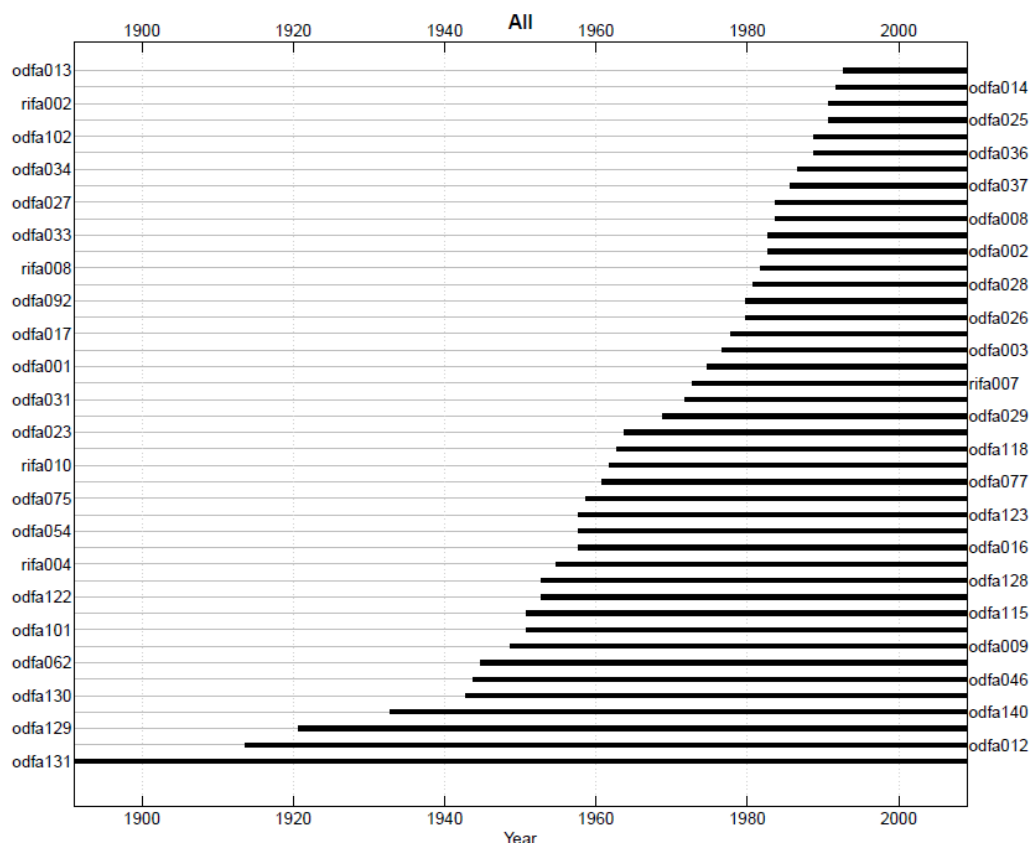


Figure 20 - Temporal length of each radius sampled from all individual tree.

The common intervals with an EPS value higher than 0.8 was chosen as the interval analysis for each group in Pearson's Correlation analysis as well as in BAI and Pointer Years (table 4). After the radii selection most of the samples had a length between 40 to 60 years and the EPS value was only acceptable ( $>0.8$ ) for the period 1967-2009.

Table 4 - EPS values for all chronologies groups

	All	Downstream	Upstream	Far	Near
Period of years	1967-2009	1979-2009	1979-2009	1974-2009	1973-2009
EPS value	0.842	0.836	0.859	0.806	0.812

The procedure for obtaining the chronology, explained before in the methods, was repeated for all study groups obtained from the original data. In total 5 chronologies were made: All trees, Upstream, Downstream, Near and Far (figures 21, 22, 23).

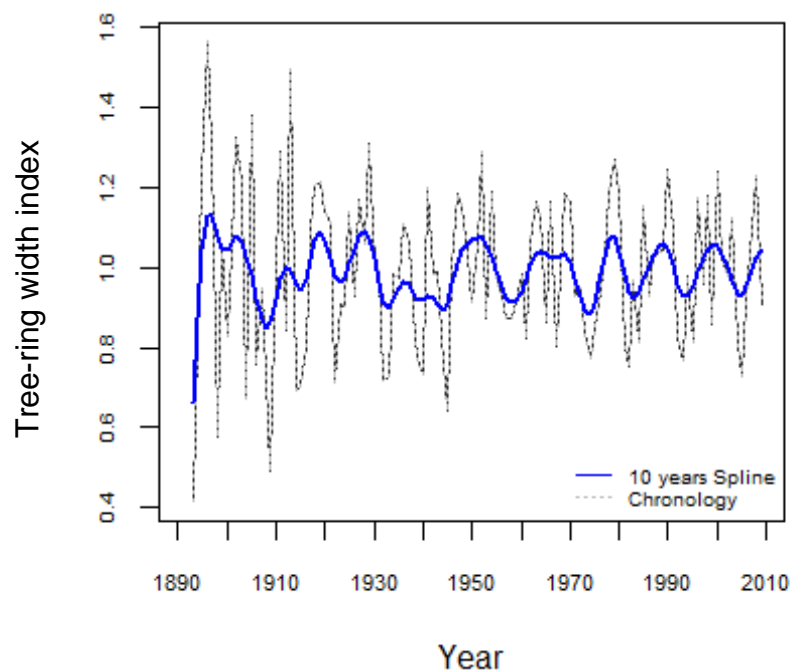


Figure 21 – Tree-ring width index for all trees.

Upstream and Downstream groups (figure 22) had a matching chronology from 1960 to 2009. Previously there were few years with a similar ring width variation, especially in 1925 and 1930 where the tendencies were opposed. In general, the equivalence of the two chronologies was disrupted in the decade of 1940 to 1950.

Upstream and Near groups (figure 23) presented a smaller maximum ring width – around 1.4 mm – while in the other chronologies it went up to 1.6 mm (figures 23 and 24). Downstream group chronology registered the biggest ring-width – with a value of 1.69 in 1942 - , and the smallest – 0.47 in 1932.

Chronology for all series registered as well a minimum width of 0.41 in 1893. General growth and decline tendencies in the 10 years spline were similar in groups Far and Near (figure 23).

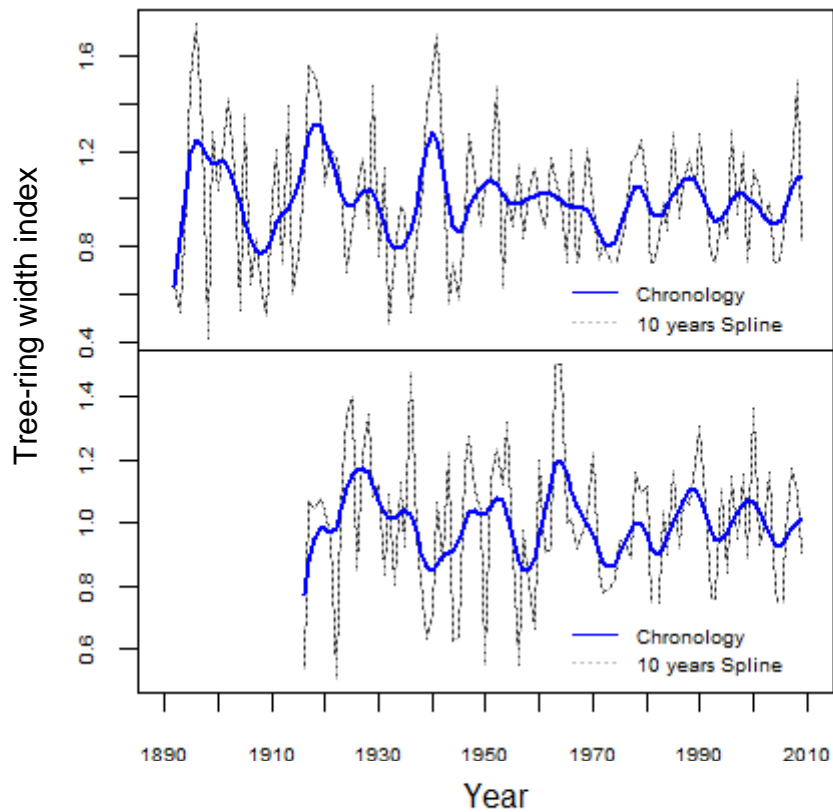


Figure 22 – Tree-ring width index all trees sampled from the Downstream (above) and Upstream (below) groups.

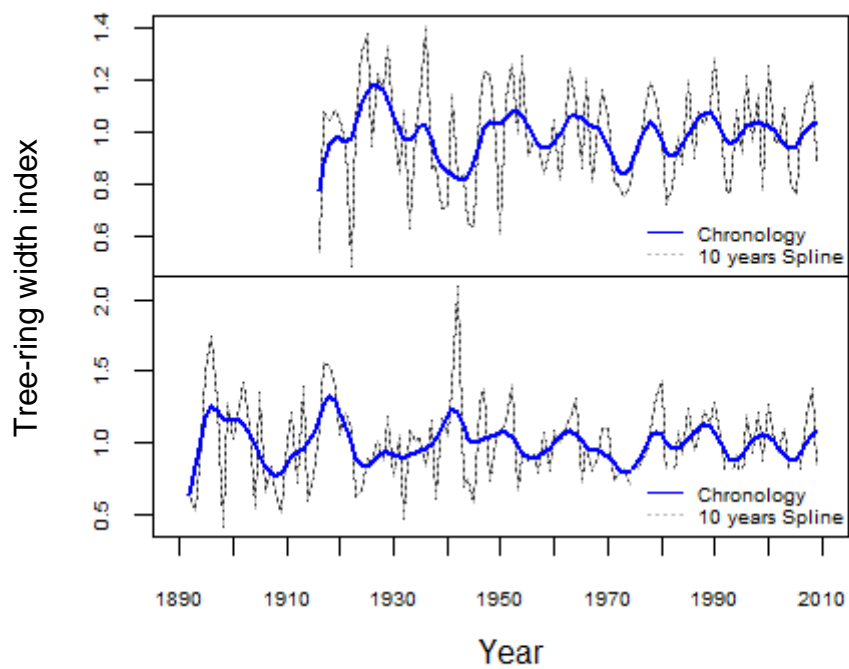


Figure 23 – Tree-ring width index for all trees sampled from the Near (above) and Far (below) groups.

#### 4.4.2 Cambial Age

Trees from Far group had a higher tree-ring width (figure 24) during the first 25 years, with a higher difference between groups Near and Far, from 5 to 12 years, of around 2mm maximum. When comparing Upstream and Downstream groups, in the first 13 years Downstream had a higher tree-ring growth, shifted for 7 years and after 20 years the two groups met and maintained the same level of tree-ring growth. During the first 7 years the difference between the two groups was bigger with a maximum of 1 mm.

In both groups there was a higher difference between tree-ring widths when trees were young, in the first 5 to 7 years of life.

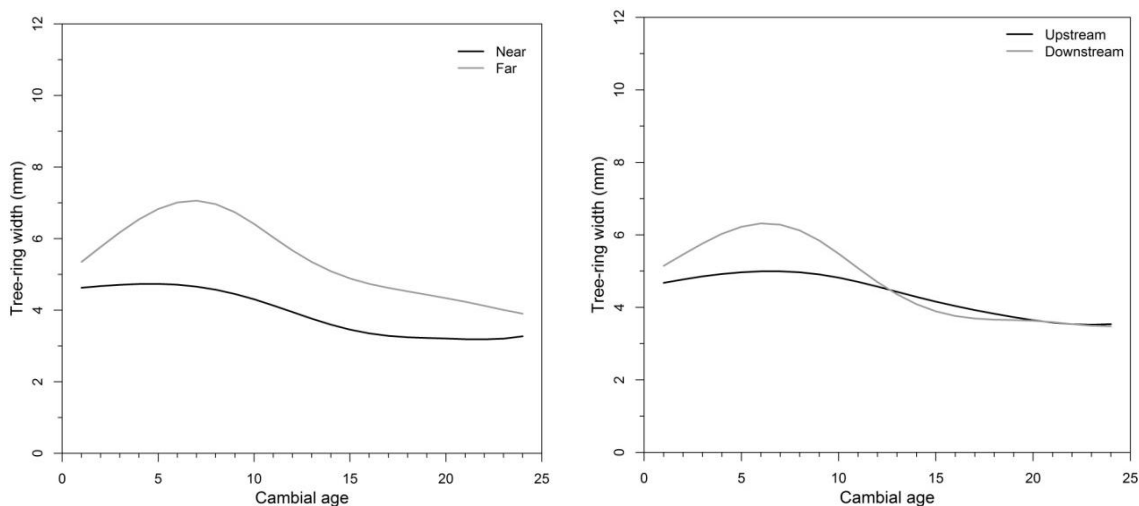


Figure 24 - Comparison between Far/Near and Downstream/Upstream cambial age groups with a 20 year spline.

#### 4.4.3 Basal Area Increment

BAI analysis was made for all groups: Far, Near, Downstream and Upstream, as well as All trees (figures 25, 26 and 27).

By visual comparison, the BAI of groups Near and Far (figure 26) had in general a similar tendency growth during the 69 years analyzed, when comparing splines. Both reached their maximum between 1990 and 2000, twenty years before there was a descending period with a significant BAI drop between 1960 and 1970. Near group had a smaller area of maximum increment (around 500 mm<sup>2</sup>) compared to Far group (reached up to almost 6800 mm<sup>2</sup>).

When examining the chronology we could see higher differences between maximum and minimum in Far group (figure 26), which happened after the recorded drought periods in Odelouca river: 1974-1976, 1980-1983, 1990-1994 and 2002-2006. These tendencies were also visible in the Near group, however it presented a smaller amplitude between minimum and

maximum in the chronology. When observing Near chronology it was visible that its peaks are homogeneous in its area values and smaller than those of Far group.

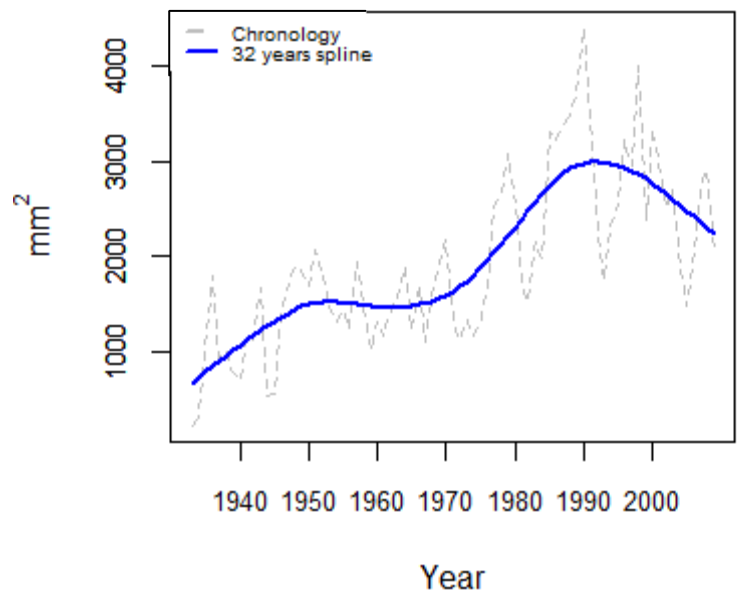


Figure 25 - BAI for All trees.

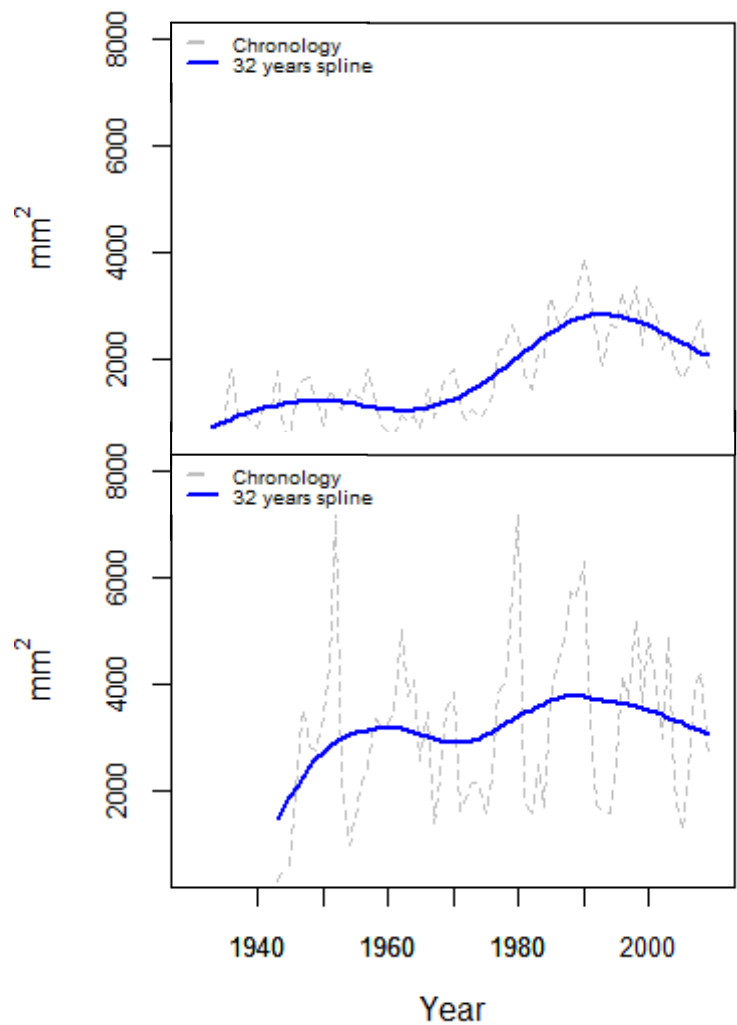


Figure 26 - BAI for groups Near (above) and Far (below).

The differences between BAI in Downstream and Upstream groups (figure 27) were more evident than the groups discussed before. In this case Downstream had a clear peak in 1990 followed by a big decrease in basal area, from 4000 mm<sup>2</sup> to 2000mm<sup>2</sup> in 20 years. The year with highest value in the Downstream chronology was the same as in the Upstream group with 2500 mm<sup>2</sup> and this value was kept constant from 1990 to 2000, showing the start of its diminishing in 2010. In general in the Upstream chronology there was a steady increase in the basal area increment with its highest variation between 1940 and 1970.

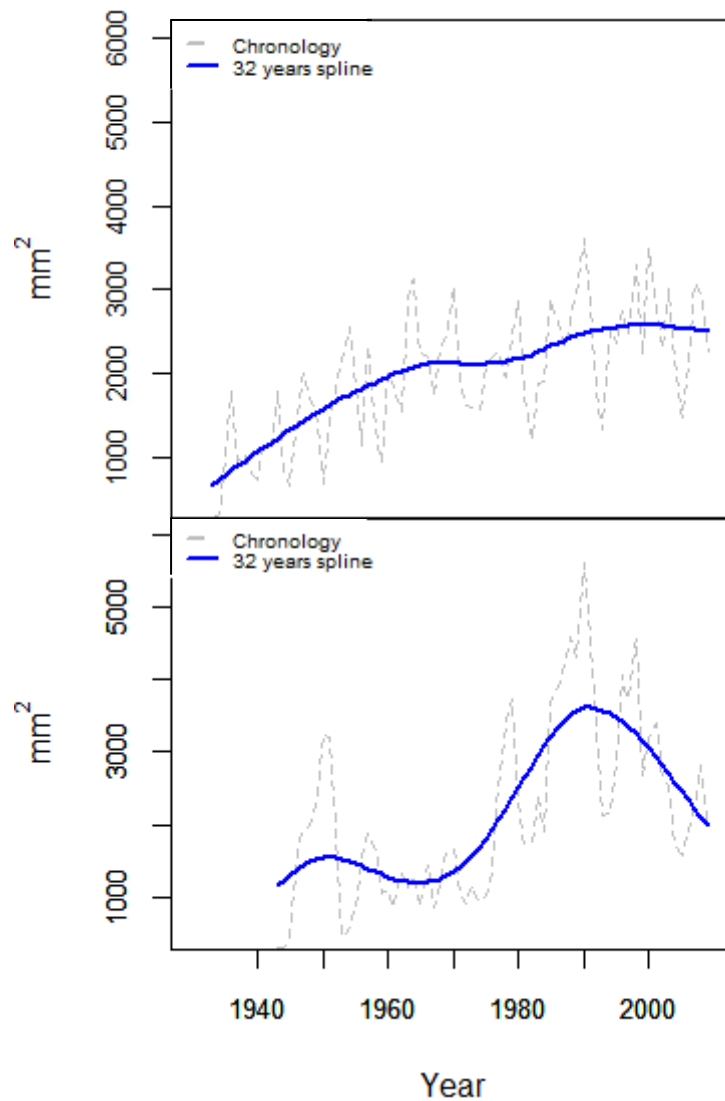


Figure 27 - BAI for groups Upstream (above) and Downstream (below).

#### 4.4.4 Pointer years

The most remarkable and consistent negative pointer year in 2005 was visible in almost all chronologies, with an exception for Downstream group (figure 30). There were a high number of negative pointer years before 1950, especially in Near, Upstream and All groups (figures 28, 29 and 30).

Upstream group had more negative pointer years than Downstream, the only pointer year they had in common was negative, in 1972. Far and Downstream groups had less negative pointer years, from all the groups analyzed. There were less positive years than negative, and they showed a higher mean growth deviation compared to any of the negative pointer years.

In the chronology for all trees there were 4 negative pointer years corresponding with drought periods: 1972, 1993, 2004 and 2005.

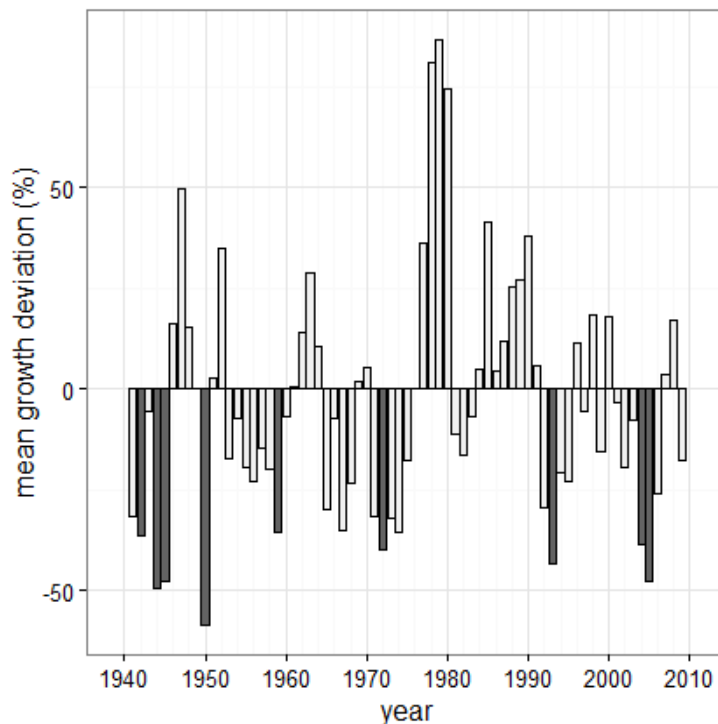


Figure 28 - Pointer years for All trees. Negative PY for 1942, 1945, 1946, 1950, 1959, 1972, 1993, 2004 and 2005.



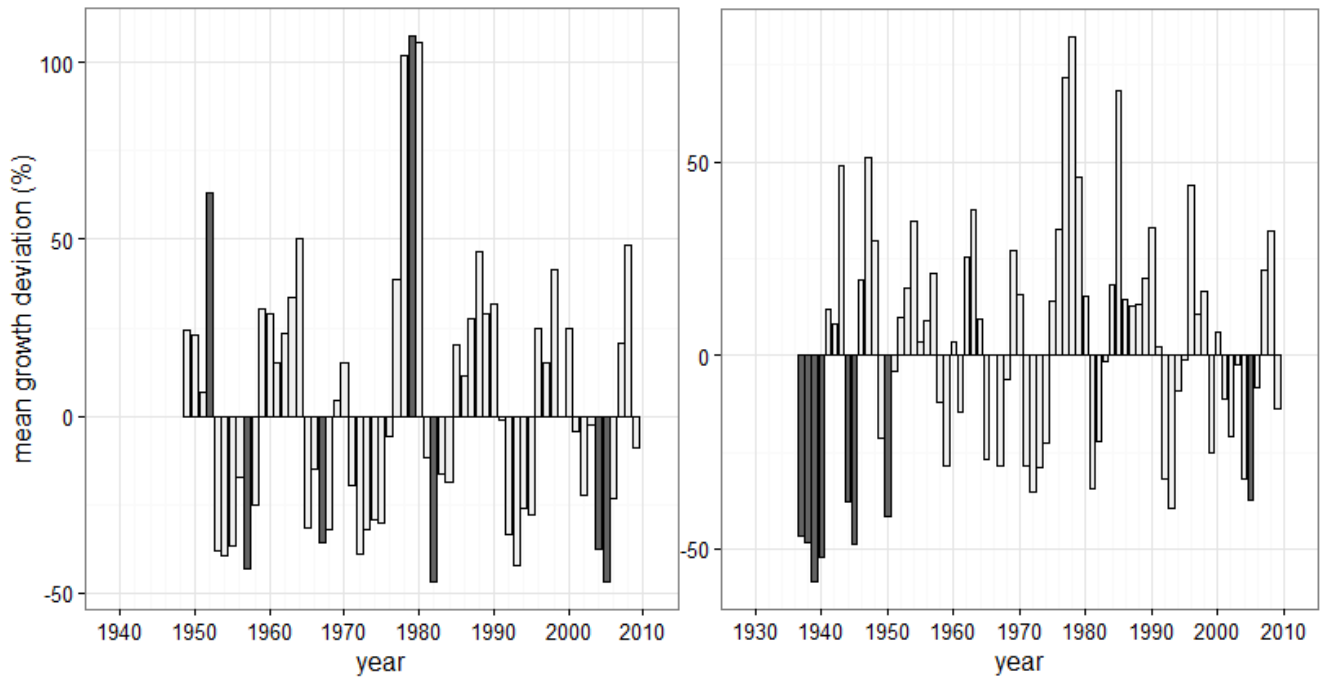


Figure 29 - Pointer years for Far (left) and Near (right) groups. Positive PY in 1952 and 1979, and negative PY in 1957, 1967, 1982, 2004 and 2005 for Far group. Negative PY in 1937, 1938, 1939, 1940 and 2005 for Near group.

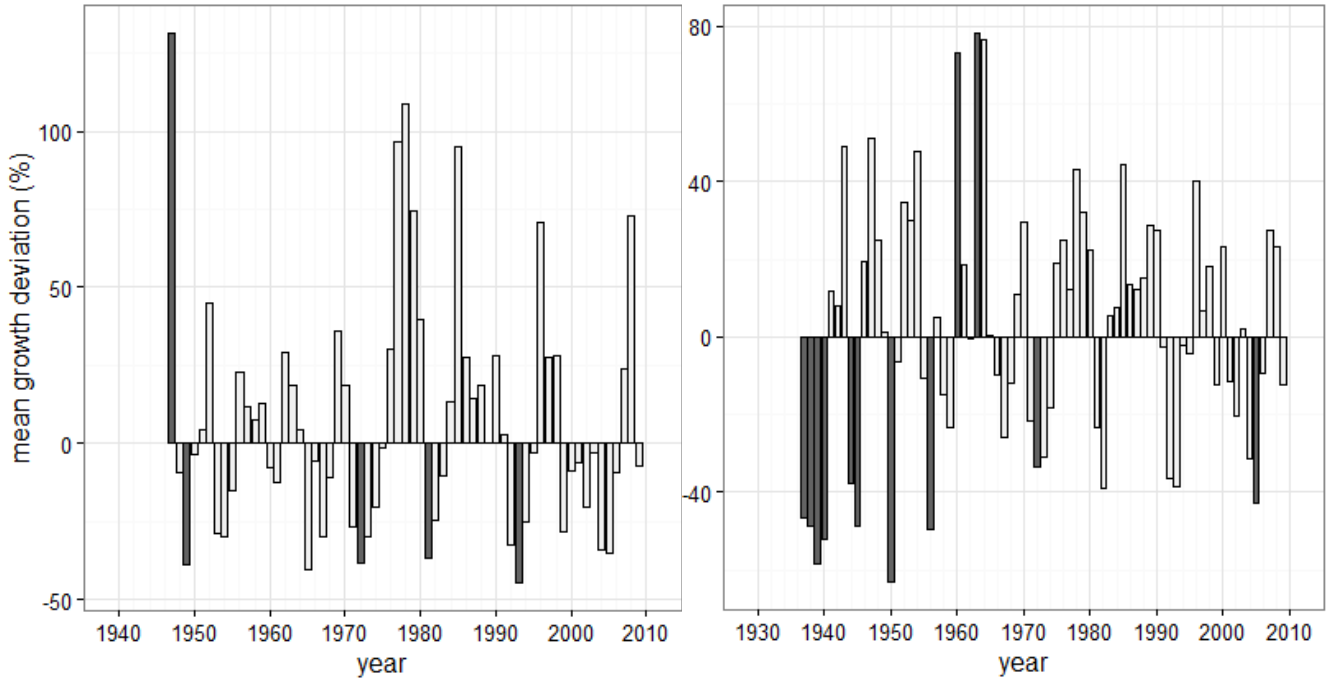


Figure 30 - Pointer years for Downstream (left) and Upstream (right) groups. Positive PY in 1947 and negative PY in 1949, 1972, 1981 and 1993 for Downstream group. Positive PY in 1960 and 1963 and negative PY in 1937, 1938, 1939, 1940, 1944, 1945, 1950, 1956, 1972 and 2005 in Upstream group.

## 4.5 Environmental factors and growth

### 4.5.1 Population response to climate and hydrology

Simple Pearson Correlations between tree-ring width, monthly total precipitation and mean temperatures (minimum, medium and maximum) and discharge were calculated. For each group a mean tree-ring width chronology was calculated with raw values, and correlations were calculated using the span years resulting from the EPS analyses.

The results for Pearson's correlations are presented below, with significant levels below  $\alpha=0.05$  shown in green when positive and red when negative. Correlation values for all groups: All, Far, Near, Downstream and Upstream (tables 5, 6, 7, 8 and 9).

Table 5 - Simple Pearson's correlations results for all trees

<i>All trees</i>	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<b>Precipitation (mm)</b>	0.152	0.129	-0.180	0.190	0.116	-0.136	0.160	-0.111	0.046	0.123	0.127	0.040
<b>Maximum Temperature (°C)</b>	0.040	0.223	0.136	0.025	-0.011	0.112	0.114	0.102	0.234	0.078	0.037	0.101
<b>Minimum Temperature (°C)</b>	0.348	0.440	0.165	0.211	0.163	0.152	0.105	0.152	0.309	0.041	-0.061	0.106
<b>Monthly Discharge (m<sup>3</sup>/s)</b>	0.457	0.414	0.148	0.472	0.468	0.620	0.497	0.130	0.009	0.168	0.233	0.301

Table 6 - Simple Pearson's correlations results for Far group

<i>Far</i>	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<b>Precipitation (mm)</b>	0.239	-0.058	-0.287	0.344	0.158	-0.034	-0.044	-0.169	-0.003	0.002	0.009	0.045
<b>Maximum Temperature (°C)</b>	0.087	0.211	0.079	0.045	-0.073	0.128	0.088	0.112	0.246	0.034	0.015	0.038
<b>Minimum Temperature (°C)</b>	0.248	0.258	0.112	0.071	0.065	0.186	0.028	0.093	0.377	-0.083	-0.028	0.036
<b>Monthly Discharge (m<sup>3</sup>/s)</b>	0.316	0.036	-0.117	0.462	0.453	0.606	0.461	0.125	-0.077	0.035	0.069	0.160

Table 7 - Simple Pearson's correlations results for Near group

<i>Near</i>	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
<b>Precipitation (mm)</b>	0.278	0.304	-0.176	0.273	0.067	-0.078	-0.032	-0.142	0.005	0.075	0.067	0.209
<b>Maximum Temperature (°C)</b>	0.021	0.231	0.161	0.016	0.029	0.087	0.132	0.115	0.222	0.112	0.026	0.114
<b>Minimum Temperature (°C)</b>	0.328	0.463	0.190	0.158	0.184	0.118	0.126	0.156	0.371	0.045	-0.056	0.169

Temperature (°C)												
Monthly Discharge (m <sup>3</sup> /s)	0.447	0.449	0.174	0.465	0.490	0.600	0.497	0.148	0.036	0.149	0.204	0.369

Table 8 - Simple Pearson's correlations results for Upstream group

<i>Upstream</i>	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Precipitation (mm)	0.296	0.222	-0.319	0.511	0.165	-0.118	-0.027	- 0.160	- 0.042	0.119	-0.012	0.206
Maximum Temperature (°C)	-0.144	0.245	0.103	-0.247	-0.041	0.068	0.142	0.194	0.127	0.080	0.027	0.035
Minimum Temperature (°C)	0.165	0.394	0.090	0.097	0.107	0.070	0.118	0.168	0.234	-0.014	-0.091	0.079
Monthly Discharge (m³/s)	0.258	0.239	0.032	0.668	0.551	0.652	0.536	0.071	- 0.123	0.262	0.191	0.314

Table 9 - Simple Pearson's correlations results for Downstream group

<i>Downstream</i>	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Precipitation (mm)	0.451	0.404	-	0.432	0.174	-0.060	0.042	-	0.016	0.032	-	0.095
Maximum Temperature (°C)	0.133	0.180	-	-0.017	-0.212	0.128	-0.058	0.058	0.006	0.028	-	-
Minimum Temperature (°C)	0.398	0.471	-	0.256	0.017	0.175	0.018	-	0.269	-0.162	-	-
Monthly Discharge (m <sup>3</sup> /s)	0.556	0.515	0.222	0.365	0.304	0.496	0.340	0.174	-0.196	0.173	0.179	0.114

There were several months with significant positive correlations with monthly discharge. These correlations happened during late spring (April to June) and winter (January and February), but in downstream group the discharge was not as important in summer months.

Precipitation had positive correlations with growth in all groups except All group (table 5), and mostly in January, February and April. Only one negative correlation was found between tree-ring width in the Upstream and precipitation in March. The highest significant correlations were in Downstream trees (table 9), for January, February and April which are just before the beginning of the main growing season. Maximum Temperature had no significant correlations in any of the groups and months.

Minimum Temperature had significant correlations with growth in all groups, in January, February and September. The effect of high minimum temperature on possible extension of growing season could be present on the tree-ring has a possible false ring, or intra-annual density fluctuations, in this case a band of earlywood would be formed within latewood (Campelo et al. 2007). In order to confirm the effect of minimum temperatures on ring-growth, each sample was observed in years with extreme temperatures in September, 2003 and 1987 (high values) and 1972 and 1974 (low values).

There was no visible trend in tree-ring appearance in the respective years in regards to size or shape of the vessels, their pattern, false rings or intra-annual density fluctuations. However, in several individuals the ring 2003 was wider than the three following years (figure 31), this change in growth was very evident, and therefore considered as unusual. This trait was present in less than 50% of the individuals, and so, not accounted as a common to the population.

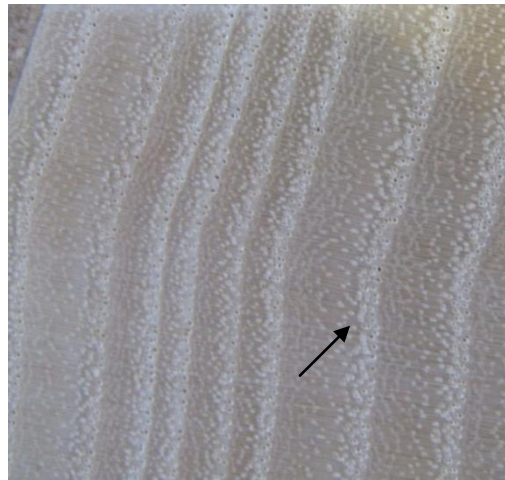


Figure 31 - Example of tree ring with unusual growth in year 2003 (pointed out with arrow) (photo by Inês Marques).

#### 4.5.2 Tree growth responses to multiple factors

The principal components circle (figure 32) identifies the variables with most interesting effects, like C/N ratio, which showed elevated relation with soil conductivity. The two variables representing fine sand ("X0.05.0.25mm") and clay and silt parts of the soil ("X0.05mm") are also related to each other and with PC2 axis.

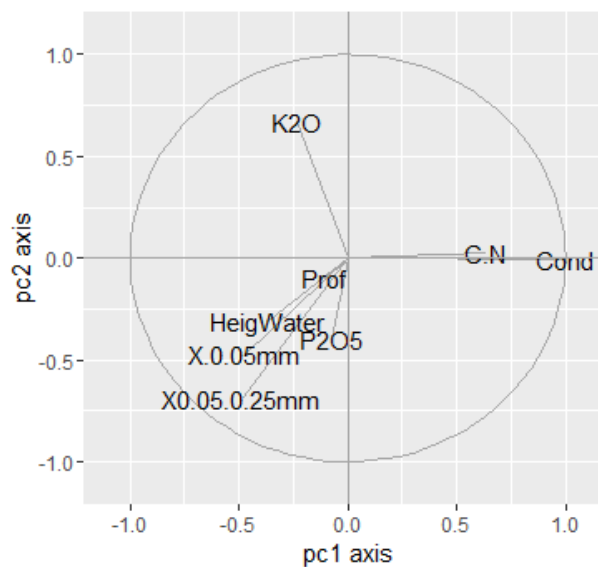


Figure 32 - PCA circle with the main variables chosen for final selection. %variance PC1 = 96.41, %variance PC2= 1.603.

Simple plotting (figure 33) between some response variables showed a clear linear relationship between some years in the chronology, height to water and phosphorous. There was less evident relation for the variables C/N ratio and fraction of the soil inferior to 0.05mm, which corresponds to clay and silt particles. After examining all possible variables, and having into account the shown figures, four explanatory variables were chosen to use in linear mixed models: Height to Water, C/N,  $P_2O_5$  and Silt and Clay soil fraction.

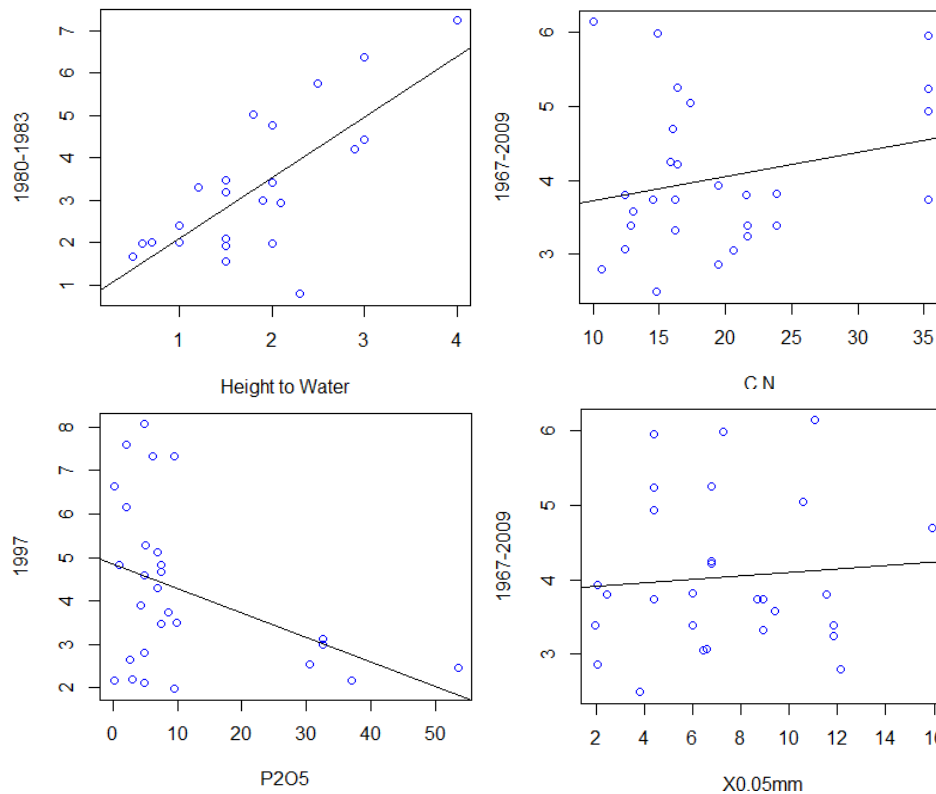


Figure 33 - Simple plotting between explanatory variables and different year periods. Height to water in *m*, C.N ratio with no units,  $P_2O_5$  in *ppm* and X0.05mm (soil particles with diameter inferior to 0.05mm) in %.

The mean growth and BAI for the periods 1967-2009, 2000-2009 and 2005-2009 were used as response variables in order to assess if there were any important factor affecting total mean growth (period 1967-2009). Because soil variables were used as explanatory variables and might only have a relation with recent events the other two periods were also analyzed (2000-2009 and 2005-2009). BAI periods were analyzed with *lme* package (Pinheiro et al. 2016) and the other periods with *glmm* package (Fournier et al. 2012; Skaug et al. 2016), both from R software (R Development Core Team 2008) considering their variables distribution.

The best fitted models, selected with AIC, for the response variables mean tree growth and BAI for 1967-2009, 2000-2009 and 2005-2009 are listed below (table 10).

Table 10 - Estimates, Standard Error (SE) and P-value for the best fitted models for all response variables

Response Variable	Fixed Effects	Model Number	Estimate	SE	P-value
1967.2009	Intercept	11	0.86775	0.12797	<0.0001
	HeigWater		0.15801	0.03761	<0.0001
	C/N		0.01203	0.00462	0.009
2000.2009	Intercept	3	0.83900	0.83900	<0.0001
	HeigWater		0.14700	0.14700	0.047
2005.2009	Intercept	3	0.68420	0.68420	<0.0001
	HeigWater		0.14750	0.14750	0.066
BAI1967.2009	Intercept	3	7.96994	0.447145	0
	HeigWater		0.19609	0.136979	0.165
BAI2000.2009	Random effect	1	4.63143	0.340242	0
BAI2005.2009	Random effect	1	4.42679	0.385015	0

Table 11 - Weight and accumulated weight values for the alternative models for mean growth

Response Variable	Model	Model Number	Weight	Accumulated Weight
1967.2009	HeigWater + C/N	11	0.37	<b>0.67</b>
	HeigWater + P <sub>2</sub> O <sub>5</sub> + C.N	17	0.15	
	HeigWater + Silt and Clay + C.N	19	0.14	
2000.2009	HeigWater	3	0.22	<b>0.64</b>
	HeigWater+C.N	11	0.13	
	Random effect	1	0.10	
	HeigWater + P <sub>2</sub> O <sub>5</sub>	10	0.09	
	HeigWater + Silt and Clay	7	0.08	
2005.2009	HeigWater	3	0.19	<b>0.60</b>
	HeigWater+C.N	11	0.16	
	Random effect	1	0.10	
	HeigWater + P <sub>2</sub> O <sub>5</sub>	10	0.08	
	HeigWater + Silt and Clay	7	0.07	

Raw mean growths showed the best fitted models for the response variables when compared to BAI mean growths. In BAI periods of 2000-2009 and 2005-2009 the best model was the one with only random effect, which means that none of the chosen variables had any influence on tree BAI mean growth. For the period of BAI 1967-2009 the best model had as fixed effect height to water, which is the same response as the models for raw tree growth for all periods.

The results of the general linear-mixed effects models indicated that mean growth for the last 42 years (period 1967-2009) increased with height to water and C/N ratio (table 10). Soil phosphorous and fine sand were not good predictors of tree growth but improved the fit (table 12). The results show that all the three alternative models included the predictor variable Height to water and accounted for 67% of the collective Akaike weight, and the best model 37% likely given the candidate set of models (table 11).

For the period 2000-2009 C/N ratio improved the fit and soil phosphorous and fine sand had a smaller Akaike weight than models exclusively with random effect. The results show that four of the five best alternative models included the predictor variable Height to water and all best models accounted for 64% of the collective Akaike weight, and the best model 22% likely given the candidate set of models. Period 2005-2009 had similar results, but with a lower collective weight, of 60% and the best model with only 19% likely given the candidate set of models.

Periods of mean growth 2000-2009 and 2005-2009 showed a positive effect with height to water but not C/N ratio and accounted for 0.19 to 0.64 of the Akaike weight, with the best model having smaller weight than 42 years mean growth's best model.

All of the best models selected with AIC for each of the intervals had height to water as the most important factor in tree growth, with a positive effect. The next best models always included height to water as one of the fixed effects, most of the times with a significant *p-value*.

Table 12 - Estimates, Standard Error (SE) and P-value of all the models for all response variables

Response Variable	Fixed Effects	Model Number	Estimate	SE	<i>P-value</i>
1967.2009	Intercept	17	0.88891	0.13585	<0.0001
	HeigWater		0.15794	0.03759	<0.0001
	C/N		0.01163	0.00469	0.01300
	P2O5		-0.00124	0.00266	0.64100
1967.2009	Intercept	18	0.87422	0.14861	<0.0001
	HeigWater		0.15916	0.03993	<0.0001
	Silt and Clay		-0.00090	0.01044	0.932
	C/N		0.01192	0.00478	0.013
2000.2009	Intercept	11	0.60097	0.27911	0.031
	HeigWater		0.17315	0.07677	0.024
	C/N		0.00976	0.00993	0.326
2000.2009	Random effect	1	1.00700	0.152	<0.0001
2000.2009	Intercept	10	0.86084	0.15945	<0.0001
	HeigWater		0.14836	0.07419	0.046

	P <sub>2</sub> O <sub>5</sub>		-0.00225	0.00533	0.674
2000.2009	Intercept		0.82185	0.17925	<0.0001
	Silt and Clay	7	0.00372	0.02200	0.866
	HeigWater		0.14152	0.08146	0.082
2005.2009	Intercept		0.36260	0.28990	0.211
	HeigWater	11	0.18020	0.08100	0.026
	C/N		0.01330	0.01040	0.199
2005.2009	Random effect	1	0.95680	0.08330	<0.0001
2005.2009	Intercept		0.72036	0.17260	<0.0001
	HeigWater	10	0.14905	0.08032	0.064
	P <sub>2</sub> O <sub>5</sub>		-0.00368	0.00570	0.519
2005.2009	Intercept		0.67327	0.19742	<0.0001
	Silt and Clay	7	0.00226	0.02318	0.922
	HeigWater		0.14441	0.08612	0.093

Intervals 2005-2009 and 2000-2009 showed no significant responses for other explanatory variables other than height to water. Both response variables had very significant p-values for random effect models.





## 5 DISCUSSION

Mediterranean climate is particular because of its seasonality, and trees in the riparian Mediterranean environment have developed adaptive responses in order to deal with water scarcity in the summer and floods in autumn and winter. The annual growth of *F. angustifolia* as a riparian species was expected to have water availability as a limiting factor in the summer and waterlogging in flood periods, like other riverine species with a similar position in the river as narrow-leaved ash (Rodríguez-González et al. 2014). Giving this, it was expected that during the drought periods, water availability (its presence/absence and the relative position of the tree in relation to the active channel) would be the main factor influencing tree growth.

This main hypothesis was supported by the presented results: water availability and tree position relative to active water channel showed to be the main factors affecting tree-ring width. Water availability impacted in several ways tree growth: discharge values in summer months, height to water and distance to river source.

Water availability in periods of scarcity showed to increase tree growth, a trend found before in *Fraxinus angustifolia* Basal Area Increment (González-Muñoz et al. 2015). In the present dissertation monthly average discharge values were most important for annual growth during summer months – April to July - which corresponded as well with the ash growing season.

However, when looking at the effect of tree position relative to water channel results from mixed models, Basal Area Increment and Cambial Age did not support the initial expectation that trees near the water channel would present higher tree-ring growth. Higher growth in group Far show that this species although having its habitat near the river, is not exclusively dependent on its discharge, but rather on other local conditions that may indirectly influence water availability within the riverine environment, like temperature, moisture, flood regime and physical soil conditions.

Indeed, height to water was the most important factor concerning position to active water channel, with a positive effect on tree-ring growth. Given the decreasing degree of flooding perturbation from the active channel outwards, higher levels on the floodplain have more developed soil, while the frequently disturbed margin are composed by substrate frequently washed by torrential foods. In Ballesteros et al.(2010) *Fraxinus angustifolia* showed less presence of injuries caused by flash-floods than trees with closer positions to the water channel, like *Alnus glutinosa* (Rodríguez-González et al. 2014). Narrow-leaved ash is considered as more tolerant to flood duration in soil than the more widely dispersed *Fraxinus excelsior* (which tolerates up to 103 days of inundation) (Ward et al. 2002). Much less comparing to a species that occurs with little to no distance from the active channel, *Salix alba* L , that tolerates up to 300 days, (Ward et al. 2002).

Together with height to water, soil features and particularly the C/N ratio, impacted *F. angustifolia* growth. The interaction of C/N ratio and climatic water balance had significant

responses in tree growth of *Fagus sylvatica*, *Quercus spp.*, *Fraxinus excelsior*, *Abies alba*, *Picea abies* and *Pinus sylvestris* in Lévesque et al. (2016), while C/N ratio alone had no significant effect. In Odelouca, C/N ratio combination with height to water improved tree ring growth response, showing a similar trend to Lévesque et al. 2016.

As a result of geomorphology dynamics of river active channel, floodplain locations have higher proportion of fine sediments, which present more C and N in the soil (Pinay et al. 1992). This soil fraction is more explored by *Fraxinus* spp (Sánchez-Pérez et al. 2008; Singer et al. 2012; Singer et al. 2014), like *Fraxinus excelsior* root system, which explores mainly fine sediment restricted to the superficial soil layers, allowing only restricted access to phreatic water levels (Sánchez-Pérez et al. 2008; Singer et al. 2012; Singer et al. 2014). Limited roots exploration to superficial horizons with fine sediment affects access to carbon, which is more present than in lower positions. A combination of root system strategy, carbon and nitrogen nutrient availability and soil composition may have resulted in higher floodplains as an evolutionary adaptive choice by narrow-leaved ash, which combined with low flow dynamics and small waterlogging periods could become a favorable environment for *Fraxinus angustifolia* growth.

Even though high flood plains show favorable conditions for this species tree growth, its position provides less access to water in drought periods. This result was evidenced by the higher variability in Basal Area Increment in Far versus Near trees. In those years the higher growth rhythm in trees farther away from water diminishes compared to previous normal hydrological years, yet regaining its normal growth rate shortly after and with higher BAI rates than Near group. Near group due to its position near to the limiting resource in drought years is able to maintain levels of growth similar to the years before the stress. Thus, trees closer to the active channel seem to have higher resistance to drought events, probably due to closest water reservoirs in the soil; on the other hand, the response observed on trees in higher floodplains suggest higher resilience to drought, given their capability of recovering from a water stress condition (Folke et al. 2004).

After discharge values in summer months and height to water, distance to river source showed also an important effect of water on tree growth. Downstream group trees had higher mean value of ring width compared to Upstream and had less significant correlations to discharge, particularly in summer months, explained by the higher discharge values. Benafátima stream was the physical separation chosen to categorize Upstream and Downstream groups, the stream flow contribution had impact on tree growth, related with higher water availability further Downstream. Not only the discharge is higher downstream but the environment is different as well, farther from the catchment, geomorphic conditions change (Rodríguez-González et al. 2014). Sediments, surface water, soil moisture and groundwater determine the vegetation that is present (Gurnell et al. 2015). These and other aspects could explain the difference between positions, and those factors should be investigated in future works.

The more significant responses for tree growth were connected to factors representing local environmental conditions, which indicates that narrow-leaved ash growth is mostly affected by site properties and not common regional signals.

When water availability or excess of it is not a limiting factor in the most problematic seasons (winter and summer), temperature plays an important role in extending the normal growing season for narrow-leaved ash. High minimum temperature in September, when growing season ends, allows an extension in growth period because the conditions (water and temperature) are maintained in a similar level to the months before. Similar trends were found in *Fraxinus angustifolia* BAI, in a riparian forest in Spain (González-Muñoz et al. 2015). Length of the growing season can affect nutrient cycle, respiration and phenological factors, already found to be related with temperature changes, especially since the 1970's (Penuelas and Filella 2001).

*Fraxinus angustifolia* has been considered as a species belonging to several functional groups, being present in disturbed rivers from frequently inundated banks to a position high and far from the active river channel (Bejarano et al. 2012). In cases of riparian gallery rehabilitation or restoration this species can be used to establish populations in areas farther away from the river but still with flood impact, tolerating waterlogging for small periods of time. Previous studies indicate narrow-leaved ash as a species that can adapt quickly and occupy areas where species less tolerant were dominant (Bejarano et al. 2012), having a clear position in the succession phase after the pioneer trees.

Taking into account the species response in a natural riverine system, as future reference, the plantation of narrow-leaved ash in rehabilitation or restoration projects, in order to be successful, should be in grounds not frequently inundated, in the areas after pioneer species establishment. *Fraxinus angustifolia* seedlings have a good ability to colonize open forest gaps or open habitats, showing vigorous growth in the first years, farther away from the active channel (Gérard et al. 2006). Its position should be in a way that it can start to occupy more evolved soil with fine sediments, due to its incapability of exploring gravel substrate, at high levels, with established forest and high canopy cover (Rivaes et al. 2010; Rivaes et al. 2013).

Although the majority of the samples had a length up to 70 years, which is not common for riverine species, the EPS value was only higher than 0.8 for a shorter period of time, only allowed the analysis of 40 of the total number of years.



## 6 CONCLUSIONS

The results shown here did support the hypothesis that water regime and availability was the main factor controlling tree growth in the riverine environment where narrow-leaved ash was present, mainly in summer months. For the study period water proximity was the main factor for tree growth with the interaction with carbon and nitrogen soil nutrients.

With this work it was possible to understand better the ecology of a species still not very studied, and so in the future be able to use it in natural engineering and restoration projects. Narrow leaved-ash must be used together with species more tolerant to floods and with optimum growth near the river active channel. Further work is required in order to understand the upper limit of height distribution of ash as well as nutrient and physical soil properties effect on shorter periods of growth.

Today climatic change affects tree growth in many natural ecosystems, and combining methods of past responses to natural systems along temperature rising trends would improve tree species response in restoration or rehabilitation projects. The rising of the minimum temperatures can change the phenology and growth rhythms of riverine trees, changing their future ecology and their use as ecosystem engineers.

In 2009, when wood samples were taken Odelouca was considered a river with natural hydrologic conditions, and for that an important reference system for tree growth responses, given that natural rivers are every year less in Europe. When using the results from this work it is important to keep in mind the size of the river and its morphological characteristics. They can be extrapolated for small Mediterranean torrential streams, with functional groups where *Fraxinus angustifolia* is present.

Further work in studying the impact of flow regime alteration in tree growth is now possible for this species in the same river, where a dam was built 7 years ago. Later comparison with tree growth in natural conditions would be able to explore growth responses to water regime and geomorphological stressors between positions in river and in active river channel.



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## **ANNEXE**





## Annex 1 – Field sheet

[illegible]

## Annex 2 – Results from physical analysis of soil samples

Codigo	>10m (%)	8-10mm (%)	8-5mm (%)	2-5mm (%)	Very Coarse Sand (2-1mm) (%)	Corse Sand (1-0,5mm) (%)	Medium Sand (0,5-0,25mm) (%)	Fine Sand (0,25-0,1mm) (%)	Very Fine Sand (0,1-0,05mm) (%)
odfa001	0.163	0.479	1.065	3.896	7.152	8.449	14.871	33.582	29.437
odfa002	0	0.210	0.777	4.937	8.530	9.705	22.648	35.007	19.229
odfa003	10.953	1.773	1.838	2.498	10.825	9.408	9.710	26.040	25.705
odfa008	0.839	0.000	0.210	1.514	2.735	3.226	8.528	40.785	41.830
odfa009	0	1.180	1.681	8.595	10.539	9.095	10.621	26.694	28.267
odfa012	0.921	0.000	0.463	1.690	2.892	9.185	32.493	33.265	20.353
odfa013	0.256	0.000	0.471	0.952	2.474	3.236	6.972	41.975	43.071
odfa014	2.304	1.220	2.059	9.625	12.794	11.103	14.035	25.368	21.138
odfa016	0	0.000	0.853	4.542	5.427	5.079	11.986	47.335	24.485
odfa023	0.306	0.470	1.145	7.349	9.483	9.419	12.636	29.251	29.633
odfa025	1.195	0.791	3.068	7.231	6.798	5.183	7.135	40.519	27.776
odfa026	2.35	0.966	1.436	7.415	10.857	18.803	36.797	14.907	5.425
odfa027	5.262	1.151	1.724	3.637	7.348	10.112	25.579	28.928	16.007
odfa028	0	0.511	1.826	10.871	11.732	13.701	19.426	21.997	19.660
odfa031	12.725	2.782	5.137	20.400	23.338	16.909	10.493	5.746	2.201
odfa033	4.593	2.965	4.140	17.423	17.777	15.324	13.942	12.110	11.551
odfa036	47.555	4.986	9.331	9.743	3.682	6.793	9.222	5.884	3.316
odfa 040	4.020	1.768	2.423	11.378	16.122	13.662	17.507	21.867	15.337
odag043	6.490	0.846	3.305	7.675	16.202	15.787	17.714	18.999	12.763
odag047	0	0.726	1.061	4.614	5.013	5.936	11.361	33.663	41.110
odag060	1.497	0.388	0.845	5.306	11.893	21.276	22.118	21.005	14.556
odfa062	0	0.153	0.302	2.526	7.017	20.659	26.963	26.090	15.962
odag069	1.919	0.742	1.601	11.006	15.399	13.532	16.105	21.571	17.375
odfa071	40.903	2.400	0.706	1.795	19.617	9.538	8.774	11.182	4.937

<b>odag079</b>	0.279	0.183	1.072	4.482	6.500	13.875	24.989	28.004	20.379
<b>odag090</b>	1.192	0.000	1.371	4.576	10.532	10.980	17.160	31.033	22.810
<b>odss100</b>	2.807	2.276	5.147	10.692	21.209	16.557	12.769	14.606	13.693
<b>odag110</b>	26.727	2.172	3.053	4.990	18.480	11.456	11.266	13.729	7.945
<b>odss126</b>	10.606	2.636	5.740	12.831	16.185	12.407	13.856	14.698	10.818

### Annex 3 – Results of chemical analysis of soil samples

Soil Sample Code	N-NH4 (mg/kg)	N-NO3 (mg/kg)	pH	Cond(μs/cm)	K <sub>2</sub> O (ppm)	P <sub>2</sub> O <sub>5</sub> (ppm)	C (%)	M.O (%)	Texture
odfa003	8.15	2.03	6.78	355.40	39	53.52	1.61	2.78	Sandy Loam
odfa008	6.34	1.22	6.28	142.60	74	4.35	1.21	2.08	Sandy Loam
odfa009	10.19	6.80	6.23	207.80	124	9.86	1.70	2.94	Sandy
odfa012	8.11	2.65	6.07	158.50	57	32.57	1.74	3.00	Sandy
odfa013	5.66	1.37	6.31	96.04	64	1.04	1.22	2.11	Sandy
odfa014	12.38	0.34	6.14	191.70	70	8.54	1.85	3.19	Loamy Sand
odfa016	7.05	0.40	6.28	102.30	66	6.99	1.22	2.10	Sandy
odfa023	6.25	0.29	6.01	109.30	54	5.01	1.41	2.42	Loamy Sand
odfa026	7.43	0.25	6.13	193.70	97	7.43	0.99	1.72	Sandy Loam
odfa028	9.40	0.33	5.93	181.90	77	7.43	2.11	3.64	Sandy Loam
odfa036	5.42	<0,05	6.79	135.40	84	9.42	1.06	1.83	Sandy
odag040	9.61	0.06	5.79	241.00	66	36.98	1.99	3.43	Loamy Sand
odag043	11.75	<0,05	6.01	388.90	72	0.16	1.74	3.00	Loamy Sand
odag047	11.60	5.43	5.89	163.40	76	3.02	1.81	3.12	Sandy
odfa062	6.61	2.40	6.15	104.70	75	6.11	1.34	2.32	Sandy
odag069	17.47	0.22	5.00	213.30	65	2.58	2.20	3.80	Sandy Loam
odag079	8.42	0.25	4.66	156.50	40	30.59	1.13	1.95	Sandy Loam
odss100	9.81	0.01	5.50	162.80	44	2.14	2.34	4.04	Clay
odag110	15.69	2.55	4.68	477.00	48	0.16	2.27	3.91	Clay
odss126	7.84	<0,05	5.33	537.60	59	4.79	2.78	4.80	Clay

**Annex 4 – Soil chemical analysis results for Far/Near and Upstream (Up) /Downstream (Down) groups**

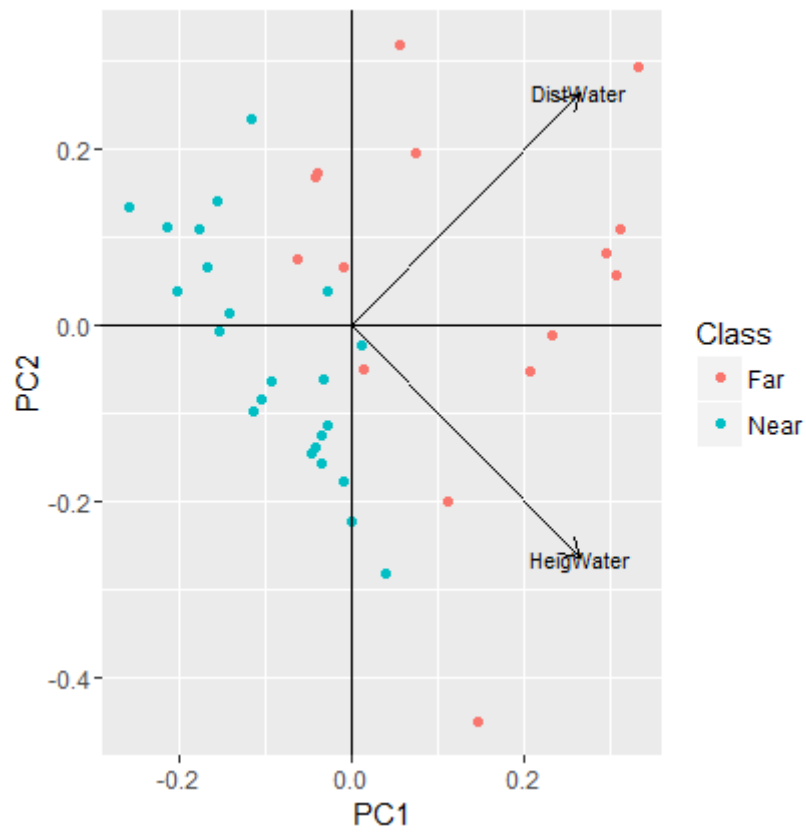
	Units	Mean		Maximum		Minimum		n	
		Far	Near	Far	Near	Far	Near	Far	Near
<b>C</b>	%	1.99	1.68	2.78	2.78	1.22	0.67	8	20
<b>O.M</b>	%	3.44	2.90	4.80	4.80	2.10	1.15	8	20
<b>NH<sub>4</sub><sup>+</sup>-N</b>	mg/Kg	7.80	9.30	10.19	17.47	5.66	5.42	8	20
<b>NH<sub>3</sub><sup>-</sup>N</b>	mg/kg	1.43	0.94	6.80	5.43	0.04	0.01	8	20
<b>Total P<sup>a</sup></b>	ppm	5.72	12.99	9.86	53.52	1.037	0.16	8	20
<b>Total K<sup>a</sup></b>	ppm	72.88	63.65	124.00	97.00	59.00	39.00	8	20
<b>pH</b>	-	5.86	5.88	6.31	6.79	5.33	4.66	8	20
<b>Conductivity</b>	µs/cm	288.19	218.40	537.60	537.60	96.04	102.30	8	20
<b>C/N</b>	-	23.27	17.84	35.29	35.29	10.01	10.63	8	20

<sup>a</sup> Total P was measured as P<sub>2</sub>O<sub>5</sub>, total K as K<sub>2</sub>O

	Units	Mean		Maximum		Minimum		n	
		Up	Down	Up	Down	Up	Down	Up	Down
<b>C</b>	%	1.35	2.18	2.11	2.78	0.67	1.13	18	13
<b>O.M</b>	%	2.32	3.75	3.64	4.80	1.15	1.95	18	13
<b>NH<sub>4</sub><sup>+</sup>-N</b>	mg/Kg	7.76	10.16	12.38	17.47	5.42	6.61	15	13
<b>NH<sub>3</sub><sup>-</sup>N</b>	mg/kg	1.28	0.86	6.80	5.43	0.04	0.01	15	13
<b>Total P<sup>a</sup></b>	ppm	13.50	7.92	53.52	36.98	1.037	0.155	15	13
<b>Total K<sup>a</sup></b>	ppm	72.67	58.92	124.00	76.00	39.00	40.00	15	13
<b>pH</b>	-	6.27	5.42	6.79	6.15	5.93	4.66	15	13
<b>Conductivity</b>	µs/cm	163.52	324.68	355.40	537.60	96.04	104.70	15	13
<b>C/N</b>	-	17.032	22.12	21.70	35.29	10.01	10.63	15	13

<sup>a</sup> Total P was measured as P<sub>2</sub>O<sub>5</sub>, total K as K<sub>2</sub>O

**Annex 5 - Principal Components correlation circle for distance and height to water. % variance PC1 = 70.79.**



## Annex 6 – Individual tree distribution by groups Far, Near, Upstream and Downstream

	Far	Near	Upstream	Downstream
odfa001	0	1	1	0
odfa002	1	0	1	0
odfa003	0	1	1	0
odfa008	0	1	1	0
odfa009	1	0	1	0
odfa012	0	1	1	0
odfa013	1	0	1	0
odfa014	0	1	1	0
odfa016	0	1	1	0
odfa017	1	0	1	0
odfa023	0	1	1	0
odfa025	1	0	1	0
odfa026	0	1	1	0
odfa027	1	0	1	0
odfa028	0	1	1	0
odfa029	1	0	1	0
odfa031	0	1	1	0
odfa033	1	0	1	0
odfa034	1	0	1	0
odfa036	0	1	1	0
odfa037	0	1	1	0
odfa046	0	1	0	1
odfa054	0	1	0	1
odfa062	1	0	0	1
odfa075	0	1	0	1
odfa077	0	1	0	1
odfa092	1	0	0	1
odfa101	0	1	0	1
odfa102	0	1	0	1
odfa115	0	1	0	1
odfa118	0	1	0	1
odfa122	1	0	0	1
odfa123	0	1	0	1
odfa128	1	0	0	1
odfa129	0	1	0	1
odfa130	1	0	0	1
odfa131	1	0	0	1
odfa140	0	1	1	0
rifa002	-	-	1	0
rifa004	1	0	1	0
rifa007	0	1	1	0
rifa008	1	0	1	0
rifa010	1	0	1	0
Total	18	24	27	16



**Annex 7 – Drainage Area, Distance to Source, Mean Annual Discharge and Mean Annual Precipitation by individual tree**

Tree Code	Drainage Area (Km <sup>2</sup> )	Distance to source (Km)	Mean Annual Discharge (m <sup>3</sup> /s)	Mean Annual Precipitation (mm)
odfa001	483.4584	47.487	2.550665	593.8463
odfa002	483.4597	47.565	2.550643	593.8396
odfa003	483.4597	47.578	2.550643	593.8396
odfa008	483.7489	47.732	2.552197	593.8512
odfa009	484.0571	47.848	2.553863	593.866
odfa012	484.4559	48.13	2.556005	593.8819
odfa013	484.5888	48.186	2.556719	593.8873
odfa014	484.6487	48.282	2.557041	593.8896
odfa016	484.6487	48.23	2.557041	593.8896
odfa017	484.6487	48.307	2.557041	593.8896
odfa023	485.4596	48.469	2.561397	593.922
odfs025	484.6894	48.369	2.55726	593.8913
odfa026	485.4775	48.506	2.561494	593.9227
odfa027	485.4799	48.53	2.561507	593.9228
odfa028	485.4967	48.59	2.561597	593.9234
odfa029	620.8578	48.583	2.561603	464.4358
odfa031	485.4967	48.614	2.561597	593.9234
odfa033	485.4979	48.634	2.561603	593.9235
odfa034	485.4991	47.22	2.56161	593.9235
odfa036	485.753	47.33	2.562974	593.9336
odfa037	485.7566	47.33	2.562993	593.9338
odfa046	539.66	49.99	2.853288	595.9838
odfa054	541.4	51.435	2.862636	596.0393
odfa062	544.1396	52.59	2.877833	596.2102
odfa075	589.5095	53.527	3.23183	615.4672
odfa077	590.0052	53.636	3.235842	615.6847
odfa092	609.0253	55.868	3.369092	620.5031
odfa101	609.6142	56.08	3.373929	620.7594
odfa102	609.6142	56.078	3.373929	620.7594
odfa115	611.8972	56.833	3.392681	621.7485
odfa118	527.3197	49.049	2.78699	595.5801
odfa122	544.1396	53.574	2.877816	596.207
odfa123	600.4513	54.078	3.29867	616.7139
odfa128	620.8925	59.361	3.466564	625.575
odfa129	620.8578	59.24	3.466279	625.5605
odfa130	616.9238	58.867	3.433967	623.9005
odfa131	620.8111	59.076	4.857068	850.4229
odfa140	484.3996	48.056	2.278404	522.5949
rifa002	278.4268	26.556	1.534965	616.3173

<b>rifa004</b>	302.6458	29.926	0.975583	359.3558
<b>rifa007</b>	302.7525	30.29	0.976459	359.5413
<b>rifa008</b>	302.6938	30.083	0.975977	359.4392
<b>rifa010</b>	302.6938	30.083	0.975977	359.4392
<b>Odelouca river</b>	712.9306	47.487	2.550665	593.8463